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MOTTLE NECROSIS OF SWEET POTATOES[1]

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INTRODUCTION

In a brief preliminary article (*10*)[2] published in 1923, mottle necrosis was first described as a distinct disease of sweet potatoes (*Ipomoea batatas* Poir.). It was pointed out that specimens collected in various parts of the United States showed in cross section brown, irregularly formed spots. Attention was called to the fact that only occasionally was an organism obtained from plantings of necrotic tissue in agar plates. A few plantings gave bacteria, a species of Penicillium, and one or two species of Fusarium but mostly *Fusarium oxysporum* Schlecht. Inoculation experiments with *F. oxysporum* gave negative results, from which it was concluded either that this organism was not the causal agent or that the proper conditions for infection had not been obtained.

In the autumn of 1924 more intensive work was undertaken, and a microscopic examination of decayed potatoes revealed the presence of a phycomycete which had not been isolated by any of the methods employed. A modification of the methods resulted in the isolation of this fungus (*7*), which was proved by numerous inoculation experiments to be the cause of the disease. Detailed investigations have since been made, the results of which are contained in this article.

HISTORY AND GEOGRAPHICAL DISTRIBUTION

Mottle necrosis, collected for the first time in 1917 on sweet potatoes of the Yellow Jersey variety at the Arlington Experiment Farm, Rosslyn, Va., has increased in importance each succeeding year. In 1918 Lauritzen collected material in eastern North Carolina on the same variety. Mottle necrosis was prevalent in New Jersey in the autumn of 1925, and farmers told the senior writer that it had occurred off and on in the State for 20 years or more.

In 1890 Halsted (*5*) described a disease of sweet potatoes in New Jersey which he called white rot. His description of white rot, probable method of infection, and other data concerning the fungus associated with it, suggest its probable identity with mottle necrosis. Halsted apparently did not study the disease in the field but called attention to the fact that, in view of the depressions about the small rootlets, infection probably occurred through them. He also pointed out that a badly decayed sweet potato might show very little evidence of decay at the surface, which is sometimes true of mottle necrosis; also that when superficially conspicuous the spots might unite and become somewhat sunken. A fungus with thick-walled

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[2] Reference is made by number (italic) to "Literature cited," p. 914.

spherical bodies, which might conceivably be an oömycete, was found associated with the disease. If one assumes from the description given by Halsted that white rot is the same as mottle necrosis, the disease has been known to science for at least 37 years.

No survey of the distribution of mottle necrosis has been attempted. Specimens have been collected in or received from Virginia, Maryland, Delaware, New Jersey, Mississippi, North Carolina, and South Carolina. Thus far it has been found mostly in States where the more susceptible varieties of sweet potatoes (Yellow Jersey and Big Stem Jersey) are grown. Only a few specimens of the disease have been received from Mississippi and no report of its prevalence there has ever been made. Whether it is widespread in the Southern States, or ever will become so, remains to be determined. Although one of the most susceptible varieties (Triumph) is grown commercially in Alabama, mottle necrosis has not been observed or reported from that State. Judging from the knowledge now available of the environmental requirements of the causal organisms, it would be surprising if it ever became very destructive in the South. There are two plausible theories to account for this: First, the varieties of sweet potato grown in the South are generally more or less resistant under field conditions, with the exception of the Triumph; and, second, results to be presented later indicate that the prevailing high temperature of the South might be a limiting factor to any general destruction of sweet potatoes in the field.

ECONOMIC IMPORTANCE

No reports of a widespread destructiveness of this disease throughout the sweet-potato-growing districts have been recorded. Losses of from 10 to 40 per cent have occurred in New Jersey and during different seasons in certain varieties grown near Rosslyn, Va. Judging from these casualties, mottle necrosis is potentially a serious disease, and, in seasons when optimum conditions for infection occur, reports of extensive losses would not be surprising.

SYMPTOMS

It is practically impossible to describe mottle necrosis in a way sufficiently accurate to differentiate it in all cases from rots caused by other fungi. If material for examination could be obtained at just the proper time and under suitable conditions it could be identified macroscopically with a reasonable degree of accuracy. On the other hand, if one is in doubt as to the identity of the disease, a microscopic examination of the decayed tissue will show the presence of mycelial threads of a phycomycete in and about the cells. Even after the organism is dead and can no longer be isolated, the large, nonseptate, vacuolated mycelial threads can be clearly distinguished. Mottle necrosis is primarily a field disease, and when it occurs it is found at digging time, being in this respect different from any of the other diseases of the roots with the possible exception of soft rot, caused by *Rhizopus nigricans* Ehrb., which may occasionally be found in the field.

Superficially, mottle necrosis may be quite inconspicuous. The surface lesions vary from small, dirty brown, somewhat sunken spots at the base of the small rootlets to conspicuous sunken spots of various

sizes and shapes. The presence of a small lesion on the surface does not necessarily indicate a small amount of decay within. Frequently such sweet potatoes may be almost entirely decayed or they may be decayed only in localized areas. On the other hand, the surface may

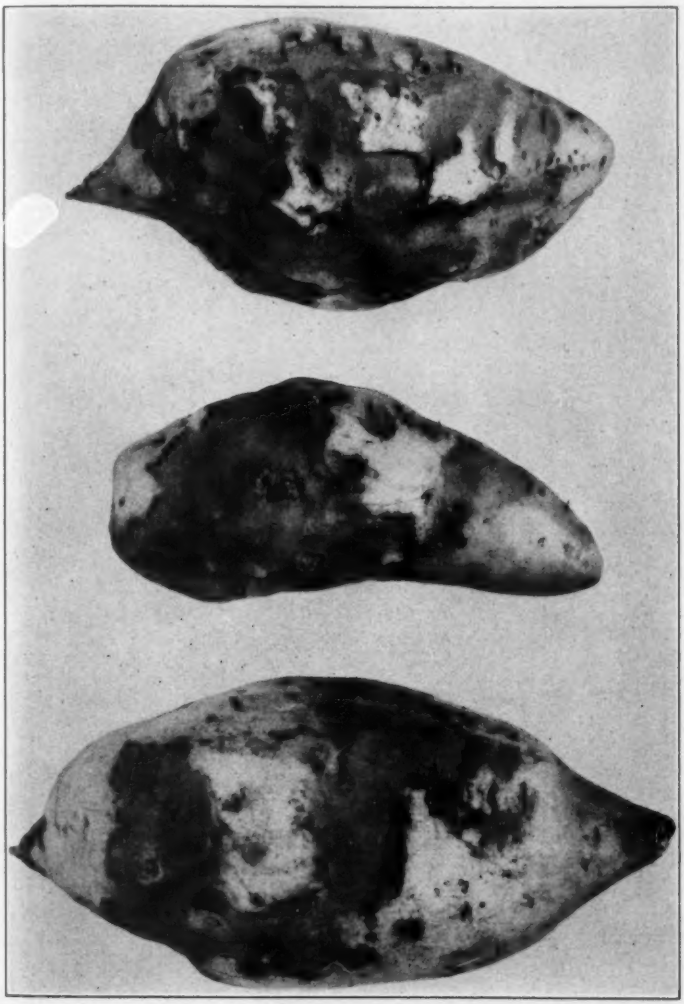

FIG. 1.—Mottle necrosis of sweet potatoes. External symptoms on the Yellow Jersey variety. (About three-fifths natural size)

be more or less covered with brown, sunken spots (fig. 1), and when the potatoes are cut open it may be found that very little internal decay has actually taken place.

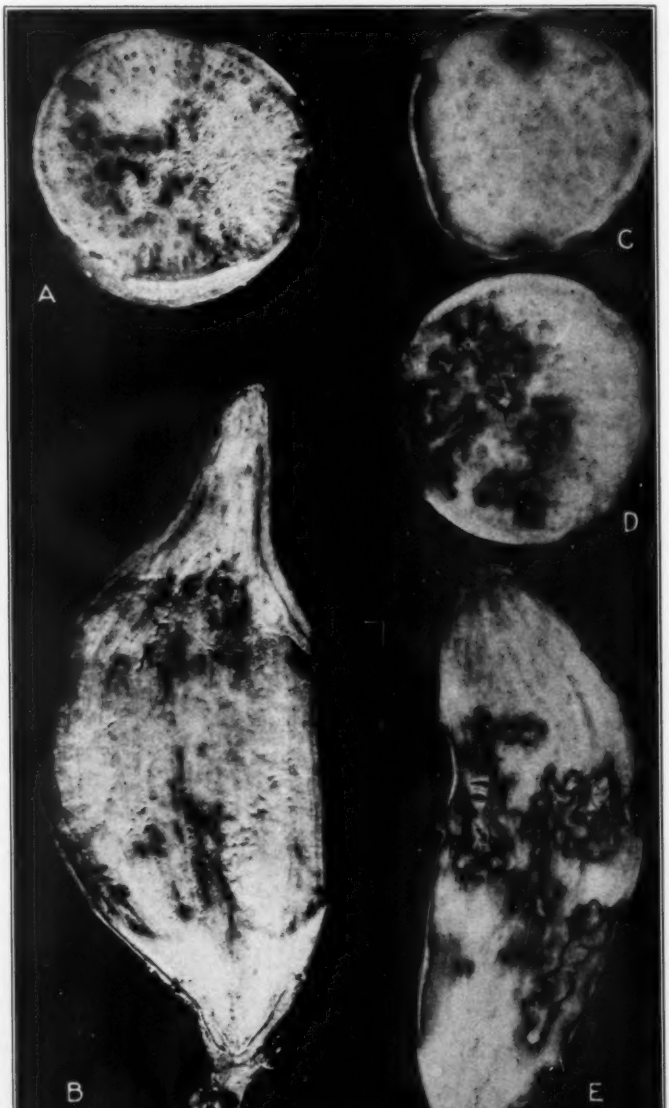

FIG. 2.—Mottle necrosis of sweet potatoes. A and B, transverse and longitudinal sections, respectively, of sweet potato, showing cheesy type of decay produced by artificial inoculation. C, transverse section of sweet potato, showing natural infection of the band type of decay. D and E, transverse and longitudinal sections, respectively, showing natural infection of marble type of decay. (All about two-thirds natural size)

When a diseased root is cut in two longitudinally (fig. 2, B and E) or transversely (fig. 2, A, C, and D), any one of three types of decay may be identified, depending to a considerable extent upon the local weather conditions which prevailed at the time decay was in progress. For the sake of classification these have been designated the marble type (fig. 2, D and E), the cheesy type (fig. 2, A and B), and the band type (fig. 2, C), the last being the least common. The band of necrotic tissue characteristic of the band type is very shallow, seldom extending beneath the fibrovascular ring, although penetration at some local point may occasionally occur. The sweet potato is firm and the chocolate-brown color of the surface is the only evidence of decay. The surface is not usually sunken, although after drying for some time (as, for example, curing in storage) some shrinking may occur. A better idea of this type of decay can be gained from a cross section through the root, which will show a thin superficial layer of necrotic tissue about 1 to 3 mm. thick.

There are well-marked characteristics that differentiate the typical marble and cheesy types of decay, both of which are encountered in the field, and, it was believed at the outset, the different symptoms were produced by different causal organisms. It was shown later that either type could be produced by more than one organism, the kind of decay depending largely upon atmospheric and soil conditions.

The marble type, shown to the best advantage by cutting a section through the root, is more prevalent in the late summer and early fall. The irregularly shaped islands of necrotic tissue of various sizes scattered more or less indiscriminately through the root are characteristic of the marble type. The organism ramifies from the point of infection in various directions, forming chambers or pockets of decayed tissue which might be assumed from a cursory examination to be disconnected. If, however, the different pockets and chambers are followed through their various ramifications, they will be found to be connected or to have a common point of origin. The necrotic tissue is dry, crumbly, and from a dark gray to a chocolate brown in color.

In a typical example of the cheesy type as found in the field or produced by artificial inoculation, the tissue is of about the consistency of soft cheese and of a grayish color. Instead of the fungus ramifying in many directions and forming almost isolated pockets, large chambers with comparatively smooth walls are produced, closely resembling those caused by Rhizopus, but less watery.

In attempting to identify this disease from a macroscopic examination, account must be taken of the conditions under which the sweet potatoes have been held. Experiments to be discussed later have shown that the color of the decayed tissue is correlated rather closely with the loss of water. If the water escapes freely the tissue will turn brown rapidly. If, on the other hand, there is very little loss of water, the color of the rotted tissue will remain practically the same as that of the healthy.

THE CAUSAL FUNGI

From the numerous inoculation experiments which have been conducted with the various strains of Pythium isolated from sweet potatoes affected with mottle necrosis, two species, identified by Drechsler, have proved to be pathogenic. The first of these, isolated

but a few times, is *Pythium scleroteichum* Drechsler (in manuscript), and is a new species to be described later by Charles Drechsler, probably in the JOURNAL OF AGRICULTURAL RESEARCH. The other species of proved pathogenicity is *P. ultimum* Trow, which was described (*17*) in 1901 as a saprophyte and is the species most frequently encountered in mottle necrosis. The present writers (*9*) have shown that this species causes a leak in artifically inoculated snap beans similar to that caused by *P. aphanidermatum* (Edson) Fitzpatrick.

The last-named organism, isolated from snap beans, has been employed in some of the investigations herein recorded and has proved to be a rapid grower and a virulent parasite on sweet potatoes, producing typical mottle necrosis.

The taxonomy of the fungi causing mottle necrosis has not been studied, the writers prefering to leave that phase of the subject to Drechsler, who is monographing the genus.

PATHOGENICITY

METHOD OF INOCULATION

Several methods of inoculation to test the pathogenicity of the organisms in question were tried before a convenient and successful one was found. A method previously used (*11*), in which the organism was grown in sweet-potato decoction for the study of the parasitism of different species of Rhizopus, was tried and found to be unsuited for Pythium. Liquid media did not, in general, give satisfactory growth, so a solid medium was eventually employed. From the different methods tried the one selected as being the most satisfactory may be described briefly as follows:

A planting is made of the organism to be used on 10 c. c. of corn-meal agar in a 90 mm. Petri dish. At the end of about two days a copious growth covers the entire plate at a temperature of approximately 25° C. Inoculations are made by what may properly be called a "well" method. A hole about 1.5 cm. deep is made into the sweet potato with a cork borer, and into the hole is placed approximately 5 sq. cm. of the corn-meal agar on which the organism is growing and the plug replaced after removing about 0.5 cm. of the lower end to allow for the inoculum. The plug forms a reasonably close seal over the agar. To prevent too rapid drying out of the culture, the plug is smeared with a thin coating of vaseline, which practically cuts off the escape of moisture. The sweet potatoes thus inoculated are placed in baskets or any covenient receptacle and held at a suitable temperature for growth. A temperature of 25° has been mostly employed for this work, although it has been shown that the optimum is somewhat lower.

INFECTION TESTS

Numerous inoculation experiments and reisolations have been made in testing the parasitism of the various species of Pythium and the susceptibility of the different varieties of sweet potatoes, all of which will not be detailed, especially those of a preliminary character. In view of the extreme susceptibility of the Yellow Jersey variety and its general distribution in the localities where mottle necrosis occurs, it has been employed for much of the inoculation work.

Literally hundreds of isolations of Pythium in pure culture have been made, many of which have been used in inoculation experiment tests, while others have never been identified. At the outset a large number of inoculations were made for the purpose of obtaining a suitable method. None of these preliminary results are included in the data shown in the tables. It is believed, however, that a sufficient number of positive results have been obtained to justify the conclusion that several species of Pythium are capable of causing a decay of sweet potatoes. All organisms except those from beans and sweet potatoes were obtained from Drechsler.

TABLE 1.—*Inoculation of different varieties of sweet potatoes with Pythium ultimum from various hosts and P. scleroteichum from sweet potatoes*

Variety inoculated	Source of organism	Pythium ultimum			Pythium scleroteichum		
		Number of potatoes inoculated	Number diseased	Percentage diseased	Number of potatoes inoculated	Number diseased	Percentage diseased
Big Stem Jersey	Sweet potato	14	9	64.3	13	7	53.8
Do	Watermelon	7	7	100.0			
Do	Pea	6	6	100.0			
Do	Pelargonium	7	7	100.0			
Nancy Hall	Sweet potato	6	5	83.3	6	3	50.0
Porto Rico	do	16	10	62.5	6	2	33.3
Triumph	do	6	6	100.0	13	13	100.0
Yellow Jersey	do	154	150	97.4	29	16	55.1
Do	Cabbage	6	6	100.0			
Do	Potato	6	6	100.0			

The data given in Table 1 show that *Pythium ultimum* and *P. scleroteichum* are parasitic on a few varieties of sweet potatoes. The former organism is commonly found on the rootlets and in potatoes affected with mottle necrosis, but the latter is only occasionally isolated. The results of the inoculations show that *P. scleroteichum* is capable of infecting several varieties of sweet potatoes, although the percentage of infections is, on the average, lower than that for either *P. ultimum* or *P. aphanidermatum*, as will be shown later. Although no difference could be detected in the morphology of the organisms from the different sources, nothing was known with respect to their comparative capacities for parasitism. The importance of this information will be appreciated if it is remembered that some of these crops (cabbage, watermelon, potato, and pea) may be, and frequently are, used in rotation with sweet potatoes. The results of the inoculations of several varieties of sweet potatoes with *P. ultimum* from five different sources in comparison with the same organism from sweet potatoes show that there is no varietal difference in the parasitism of the various strains.

The percentage of infection by *Pythium scleroteichum*, as shown in Table 1, is considerably less than that by *P. ultimum* and probably represents fairly accurately the comparative parasitic capacities of the two organisms. *P. ultimum* is a much more vigorous grower in pure culture than *P. scleroteichum* and causes on the whole a more rapid decay. At one time it was suspected that the latter produces only the marble type and the former the cheesy type, but such is not the case, since either form of decay may be produced by either species provided the environmental conditions are favorable.

VARIETY TEST

Investigations by Braun (*1*, *2*), Drechsler (*3*, *4*), Hawkins (*12*), Jones (*14*), and others (*9*, *15*, *16*) have shown that Pythiums occur on a variety of hosts widely separated in relationship; so to find them on the roots of sweet potatoes occasioned no surprise. In 1925 Harter (*6*) pointed out that the root tips of sweet potatoes in the hotbed were conspicuously injured by a species of Pythium of the debaryanum type. Out of the 21 varieties examined all were injured, but some to a greater extent than others. In view of these results, it is likely that all varieties of sweet potatoes are potential hosts to species of Pythium.

Field observation among 21 varieties of sweet potatoes grown near Rosslyn, Va., showed that the Triumph, Yellow Jersey, and Big Stem Jersey are the most susceptible varieties, although infections of some of the others were observed occasionally. Experiments have shown that the species of Pythium causing mottle necrosis is identical with the one almost uniformly isolated from the root tips of plants in the hotbed. Inasmuch as mottle necrosis is uncommon on most of the varieties of sweet potatoes under field conditions, a test of the susceptibility of 21 varieties, many of which are grown extensively commercially, was made with *Pythium ultimum* and *P. aphanidermatum*. These inoculations were made with sweet potatoes which had been held in storage for several weeks, previous investigation having shown that they may be successfully infected at any age.

TABLE 2.—*Results of inoculation of different varieties of sweet potatoes with Pythium ultimum and P. aphanidermatum*

Variety inoculated	Pythium ultimum			Pythium aphanidermatum		
	Number of potatoes inoculated	Number diseased	Percentage diseased	Number of potatoes inoculated	Number diseased	Percentage diseased
Big Stem Jersey	14	13	92.8	6	6	100.0
Creola	6	6	100.0	6	6	100.0
Dahomey	6	6	100.0	6	6	100.0
Dooley	6	5	83.3	6	2	33.3
General Grant Vineless	6	6	100.0	6	6	100.0
Georgia	9	9	100.0	8	8	100.0
Gold Skin	6	6	100.0	6	6	100.0
Haiti	6	6	100.0	6	6	100.0
Key West	6	6	100 0	6	6	100.0
Nancy Hall	6	5	83.3	6	6	100.0
Pierson	6	6	100.0	6	6	100.0
Porto Rico	16	9	56.3	16	10	62.5
Pumpkin	6	4	66.7	6	6	100.0
Red Brazil	6	6	100.0	6	6	100.0
Red Jersey	6	6	100.0	6	6	100.0
Southern Queen	6	6	100.0	6	6	100.0
Triumph	6	6	100.0	6	6	100.0
White Yam	6	6	100.0	6	6	100.0
Yellow Belmont	6	6	100.0	6	6	100.0
Yellow Strasburg	6	6	100.0	6	6	100.0
Yellow Jersey	[a]182	161	88.5	6	6	100.0

[a] Includes potatoes used in other experiments.

Table 2 shows, first, that with a few exceptions all the varieties tried were 100 per cent infected with *Pythium ultimum* and *P. aphani-*

dermatum; second, that some of the varieties never found infected under field conditions were 100 per cent infected artificially; and third, that some of the susceptible varieties (Yellow Jersey, Big Stem Jersey) under field conditions were not all infected. This is not to be interpreted, however, as indicative of any resistance on the part of such varieties, but rather of their having escaped infection. More varieties were 100 per cent infected with *P. aphanidermatum*, the species from beans, than with *P. ultimum*, the sweet-potato form.

RESULTS WITH DIFFERENT SPECIES OF PYTHIUM

The data so far recorded are the results of inoculations with only three species of Pythium, of which two (*P. ultimum* and *P. scleroteichum*) were isolated from sweet potatoes and the other (*P. aphanidermatum*) from beans. Other species of Pythium occur on the roots or other parts of plants some of which may be and frequently are used in crop rotation with sweet potatoes. Inasmuch as the sweet potato might be exposed to possible infection by such species, several of them were obtained from Drechsler and a few sweet potatoes (Yellow Jersey) were inoculated by the usual method. These organisms were compared in parallel experiments with those obtained by the writers from sweet potatoes and beans.

Out of the many isolations made from the rootlets and enlarged roots of sweet potatoes, occasionally an undetermined species of Phytophthora was obtained. While it was suspected that this organism had no causal relationship to mottle necrosis, a number of inoculations were made, the results of which will be set alongside those with the different species of Pythium.

TABLE 3.—*Results of inoculating sweet potatoes (Yellow Jersey) with different species of Pythium and with Phytophthora sp.*

Species	Host	Number of potatoes inoculated	Number diseased	Percentage diseased
Pythium:				
Adhaerens *	Cucumber	12	0	0.0
Aphanidermatum	Bean	6	6	100.0
Artotrogus	Pea	32	0	0.0
Complectens	Pelargonium	12	0	0.0
Species 5825	Pear	12	1	8.3
Debaryanum	Sweet potato	24	19	79.1
Megalacanthum	Lettuce	12	0	0.0
Splendens	Pelargonium	12	11	91.7
Ultimum	Sweet potato	82	80	97.6
Scleroteichum	do	29	16	55.1
Phytophthora sp	do	48	0	0.0

* This is a new species to be described by Drechsler later, probably in the Journal of Agricultural Research.

The data in Table 3 show that neither mottle necrosis nor any other type of disease of sweet potatoes is caused by the species of Phytophthora tested. It is probable that this organism, like many others, finds its way into the roots as a secondary invader and can in no way be assumed to bear any causal relationship. With respect to the other organisms, the results indicate that five species of Pythium may cause decay of sweet potatoes by the methods of inoculation employed. The question naturally arises, and especially in the case

of some of the Pythium species, as to what actually constitutes infection. The results in the case of some of the species are so striking as to leave no doubt. For example, *P. ultimum*, *P. debaryanum* Hesse, and *P. aphanidermatum* produce a characteristic and abundant rot. Contrasted with these unquestionably parasitic species are those which cause absolutely no decay and would unhesitatingly be classed as nonparasitic. There is in Table 3 a third group, consisting of two species (*P. splendens* Braun and *P.* 5825), which have been classed as parasites, about which there is some question. The method employed in the inoculation was the usual one in which a plug was removed from the sweet potatoes by means of a cork borer, the inoculum put in the hole, and the plug replaced. Naturally, some of the cells adjacent to the cut made by the cork borer would be mutilated and therefore would be unable to offer the same resistance to invasion by fungi as the unwounded ones. Both *P. splendens* and *P.* 5825 caused the formation of a narrow fringe of necrotic tissue adjacent to the cut surface in contact with the inoculum. In no case was a measurable amount of dead tissue produced. There was more, however, than occurred in the controls receiving similar mechanical injury but no inoculum. For this reason the writers have placed these two species in the parasitic class, though it must be kept in mind that they are extremely weak parasites on sweet potatoes.

SUMMARY OF INOCULATION EXPERIMENTS

Out of hundreds of isolations from decayed rootlets and mottle-necrosis sweet potatoes, two species of Pythium have predominated, *P. ultimum* and *P. scleroteichum*. Both of these will produce typical symptoms of mottle necrosis of the marble or the cheesy type. A species of Phytophthora was isolated several times, but in no case did it cause a decay as a result of inoculation experiments. Another species of Pythium, *P. aphanidermatum*, isolated from beans, was found to be extremely parasitic on sweet potatoes. This species, together with *P. ultimum* and *P. scleroteichum* and the very weakly parasitic forms, *P.* 5825 and *P. splendens*, must be regarded as the only species parasitic on sweet potatoes known at the present time. Under field conditions only three varieties of sweet potatoes (Triumph, Yellow Jersey, and Big Stem Jersey) have shown any marked susceptibility to mottle necrosis. However, inoculation experiments of sweet potatoes from storage have shown that all the 21 varieties tried can be successfully infected. In view of this fact, most if not all of the commercial varieties may be regarded as potentially susceptible to mottle necrosis.

DISSEMINATION

So far as known, there is nothing distinctive with respect to dissemination of the causal organism. Pythiums are known to be widely distributed in the soil and on plant parts, so their presence might be expected in almost any situation. A rootlet rot caused by Pythium occurs in the hotbed on all varieties, and, although it is present every year, it is worse some seasons than others, the prevalence of the disease probably depending upon environmental con-

ditions. Rootlet rot occurs in beds prepared by the use of sand and also in soils containing a considerable quantity of organic matter in the form of stable manure. The disease, therefore, probably is carried to the field on the plants. This, however, does not necessarily mean that because plants with diseased rootlets are planted mottle necrosis will occur in the field.

If conditions are favorable, the repeated use of the same hotbed soil appears to increase the amount of rootlet rot; also repeated planting in the same ground results in increased mottle necrosis in the field.

A certain relationship seems to exist between the soil type and mottle necrosis, although this phase of the problem has not been thoroughly investigated. As proof of this it may be pointed out that when the Yellow Jersey variety of sweet potato is planted across a field containing a loam, a sandy loam, and a very light sandy soil, the disease, while occurring on all three, is more prevalent on the sandy loam, showing that very heavy and very light soils are more or less unfavorable to the disease. To what extent soil moisture is a factor in infection has not been determined, although it is observed that during dry seasons infections are deeper in the ground than in wet ones, the moister earth seemingly contributing to infection. It is likely that an amount of soil moisture sufficient to permit zoöspore migration and mycelial development would somewhat facilitate infection.

METHOD OF INFECTION

Inasmuch as infection takes place underground, it is impossible to acquire any exact information as to just how it is accomplished. *Pythium ultimum* or *P. scleroteichum* has been isolated from the small rootlets on the fleshy roots connecting with cavities or chambers of necrotic tissue within the potato. Reasoning from these observations and from the results of the isolation of parasitic species of Pythium from the short dead rootlets and from the cavities of necrotic tissue connecting with them, there would seem to be little doubt that the small rootlets are the primary infection courts.

EFFECT OF THE FUNGUS ON THE HOST

A sweet potato freshly decayed with Pythium under favorable environmental conditions is found, on cutting in two, to be spongy or of about the consistency of a soft cheese. In the early stages of decay the color is very little changed, but upon exposure to the air it darkens, simulating old decayed tissue. The sponginess of the decayed tissue is doubtless due to the loss of turgidity caused by the death of the cells accompanied or followed by the dissolution of the middle lamellae. No studies have been made of the enzymes secreted by Pythium, but reasoning from what is known of the enzymic activities of Rhizopus spp. (*8*) causing soft rot of sweet potatoes, it is probably safe to assume that an enzyme, pectinase, is secreted which dissolves the middle lamellae in advance of the growing hyphae. Some justification for this assumption may be claimed in view of the fact that, as in the case of Rhizopus, there is a zone of decayed tissue adjacent to the healthy from which Pythium can not be isolated.

A clear picture of the relationship existing between the host and the parasite may be obtained from the study of material embedded in paraffin, sectioned, and stained with Delafield's haematoxylin and safranin. The fungus threads stain dark so that the outline of the hyphae and the granular contents of the mycelium are clearly pictured. The cell walls of the host also stain dark, but they are so much thinner than the mycelium that there is no possibility of confusing them. The cell contents stain a bright red. A careful examination of such preparations reveals the tissue composed of disconnected, collapsed cells, probably due in part at least to the loss of turgidity as a result of their death. The fungus invades the intercellular spaces and penetrates the cell wall, accumulating in the cells in considerable mass.

It is not known whether the cell wall is penetrated by mechanical pressure, by means of a dissolving enzyme, or by a combination of both. Hawkins and Harvey (*13*) seemed to be of the opinion that *Pythium debaryanum*, the cause of potato (*Solanum tuberosum* Linn.) leak, penetrated the cell wall by mechanical pressure and that no enzyme was associated in the process. In mottle necrosis of sweet potatoes the hyphae are greatly constricted at the point where they pass through the cell wall but after penetration enlarge to their original size. This fact further bears out the supposition that mechanical pressure is the method of cell-wall penetration, but does not necessarily preclude the possibility of enzymatic action. On this point Hawkins and Harvey (*13*) say: "If the fungus secretes an enzym which acts on the cell wall, it would seem probable that this enzym action would continue after the tip of the hypha passed through and the opening would be larger than the hypha. . . . With *Pythium debaryanum* on potato, however, the opening in the cell wall is never larger than the mean diameter of the hypha in the lumen of the cells and is usually considerably smaller."

There is nothing peculiar or distinctive about the mycelium within the cell. The hyphae usually enter the cell at right angles to the wall and continue in the same direction after penetration until they reach the opposite wall. Some of the mycelial threads, however, follow the inner wall and remain in close contact with it. In no case have hyphae been observed in contact with starch grains.

PHYSIOLOGICAL STUDIES

EFFECT OF TEMPERATURE ON MYCELIAL GROWTH

Because of certain irregularities in the growth in pure culture of *Pythium aphanidermatum*, *P. ultimum*, and *P. scleroteichum*, the three species with which most of the investigations were made, it was suspected that they possessed different temperature relations. This was particularly true of *P. scleroteichum*, which made a rather feeble growth at temperatures where *P. aphanidermatum* and *P. ultimum* thrived. A comparison of the growth of the three species on corn-meal agar in Petri dishes was therefore undertaken.

The methods employed are briefly as follows: Incubators with a range of temperatures varying from 2° to 41.8° C. and constant within 0.5° were employed. Ten cubic centimeters of corn-meal agar was poured into 9 cm. plates and distributed uniformly by

tilting from side to side while the agar was still hot. Plates with a thin covering of agar at the center were discarded. As soon as the medium had hardened a section 7 mm. in diameter was cut from a 2-day-old plate culture and placed at the center of the new plate. Five or more such plates were prepared for each organism for each temperature. The Petri dishes were inclosed within a covered moist chamber in order to prevent contamination with foreign organisms, of which there was very little within the duration of the experiments. In all cases where growth was interfered with by contamination the plates were discarded.

Pythium grows so rapidly that a Petri dish is often covered in less than 48 hours at the optimum temperatures. In view of this fact only two measurements were made and the optimum for growth

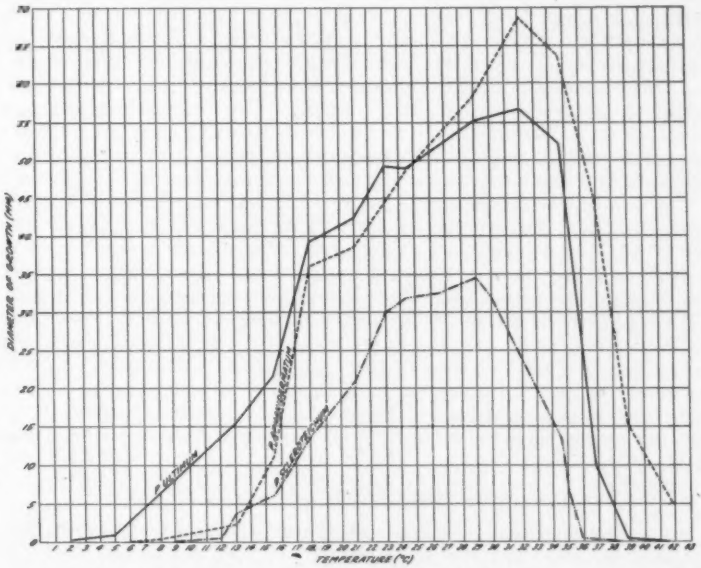

FIG. 3.—Influence of temperature at end of 24 hours on mycelial development of three parasitic species of Pythium grown in pure culture on corn-meal agar. (Measurements less than 1 mm. are estimates)

was assumed to be the temperature at which the greatest diameter of growth took place in 24 hours. The growth at the different temperatures is shown in Figure 3, the temperatures being plotted on the ordinate and the average diameter of the colonies in millimeters on the abscissa.

A longer exposure to the different temperatures was necessary to establish the maximum and minimum for growth. The maximum and minimum were considered to be those extreme temperatures at which only a slight amount of growth took place. The question of time is, of course, an important factor in establishing the extremes, and particularly so at the lower temperatures. However, it was found that a temperature 1° to 2° C. above the maximum would kill the organism within a few days. At the lower temperatures

the plates were exposed for about six weeks, and, as would be expected, none of the organisms were killed. The optimum temperature for growth of *Pythium aphanidermatum* and *P. ultimum* was found to be 32° and of *P. scleroteichum* 29° (fig. 3). The highest temperature employed was 41.8°, where all the organisms were killed in 24 hours. *P. aphanidermatum* made a growth of 15.5 mm. at a temperature of 38.8°, while *P. scleroteichum* was killed and *P. ultimum* made scarcely any growth. The maximum for growth of *P. aphanidermatum* is probably slightly above 41.8°. While neither *P. ultimum* nor *P. scleroteichum* was killed at 36.8°, the growth was very slight, the former growing only 10 mm. and the latter just starting in 24 hours. These results indicate that a temperature of 36.8° was practically the limit for *P. scleroteichum;* that for *P. ultimum* was slightly higher. It is interesting to note in connection with the maximum temperature that a visible growth on the plate can not be interpreted strictly to mean that the organism will endure such temperatures. The death of the fungus at a killing temperature is not instantaneous. Pythiums grow very rapidly, and some growth would result before death would occur. The only safe criterion to follow is to determine if the organism will make additional growth when removed to a more favorable temperature. This method was followed, and the conclusions are drawn from the results so obtained.

The minimum temperature for growth was obtained by exposing cultures of the different species for 39 days at temperatures of 2.5° and 8° C. As a result it was found that in no case were any of the organisms killed, as was shown by their rapid growth when removed to a warmer temperature. *Pythium aphanidermatum,* with a maximum higher than either of the other species, has a minimum of 6°; *P. ultimum,* 2°; *P. scleroteichum,* about 8°. *P. scleroteichum,* with an optimum (29°) 3° lower than the other two species, has the narrowest range of growth, that of *P. ultimum* and *P. aphanidermatum* being about the same. The greatest diameter of growth over a 24-hour period was produced by *P. aphanidermatum* and the least by *P. scleroteichum* (fig. 3).

INFLUENCE OF TEMPERATURE ON AMOUNT OF DECAY

Investigations have shown that the optimum for the growth of *Pythium ultimum* is approximately 32° C., the minimum about 2°, and the maximum about 36.8°. A priori, it might be assumed that the optimum for the growth of the organism in pure culture would be approximately the same as that which would give the maximum amount of decay in a given time. This, however, appears not to be the case, at least so far as *P. ultimum* is concerned. As this organism is most frequently isolated from mottle-necrosis sweet potatoes, it was used in these investigations.

A preliminary experiment was made with the Triumph variety in order to acquire some information with respect to the probable location of the critical temperatures. Ninety-six sweet potatoes of this variety were inoculated in the usual manner, apportioned into 6 lots, and held for 5 days in incubators, the temperatures of which ranged from 5° to 35° C. At the end of this time the potatoes were removed from the incubators, cut open longitudinally, and measurements made of the spread of decay. The results are given in Figure 4

and show that the spread of the decay increases with the rise in temperature up to 12°. Above this temperature the amount of decay decreases up to about 35°, where there is merely a trace of rotting. In another experiment, 20 inoculated sweet potatoes of the Triumph variety were held at 38° for 5 days, and no signs of decay appeared (fig. 4). From these data it may be concluded that *Pythium ultimum* may infect the Triumph variety over a wide range of temperatures (5–35°), with a minimum and a maximum slightly below and above those figures, respectively, and an optimum of about 12°.

As the available supply of the Triumph variety was insufficient, and as its normal shape—fairly long and cylindrical—did not lend itself readily to quantitative study, it was deemed best to change to the Yellow Jersey, a variety which is normally fusiform to globular or ovoid. The experiments with the Triumph variety, however,

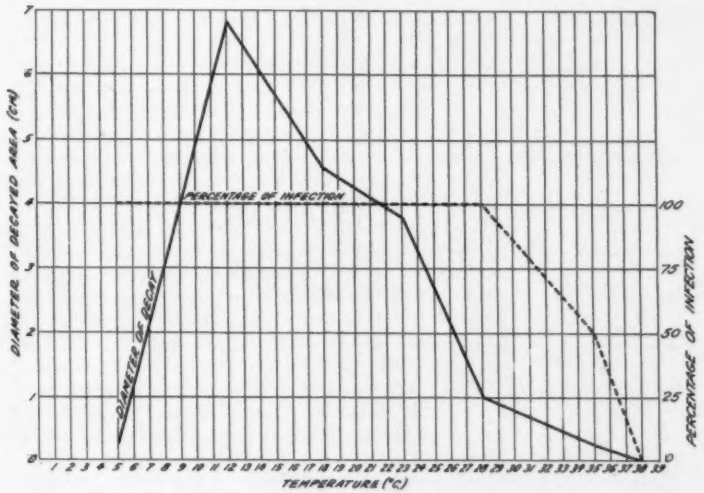

FIG. 4.—Influence of different temperatures on diameter of decayed area and percentage of infection of Triumph sweet potato inoculated with *Pythium ultimum*, after five days

served as a guide in the setting up and carrying out of later experiments. All the conclusions drawn and the data used in plotting Figure 5 are taken from two parallel sets of corroborating experiments made with the Yellow Jersey variety.

The experiments on the amount of decay were conducted as follows: Fairly large chunky sweet potatoes of as near uniform size as possible were selected and then cleaned by washing in water, after which they were inoculated by the same method as that employed in the pathogenicity tests, except that the well was made a little deeper in order that the inoculum might be placed as near the center of the sweet potato as possible. Ten sweet potatoes so treated were placed in wire baskets and exposed to temperatures ranging from 2° to 36° C. The duration of the experiment was three days, at the end of which time the potatoes were removed and weighed. They were then cut open, the decayed tissue was removed, and the healthy portion

weighed. The amount of decay from all the sweet potatoes at a given temperature was used as a basis for computation. The temperature at which the greatest total amount of decay occurred was regarded as the optimum temperature. To determine the maximum and minimum temperatures, separate experiments were set up and the time of exposure was increased; at the lower temperature the time was increased to several weeks. At the higher temperature decay resulted within a few days or not at all, or the sweet potato was rotted by some other organism.

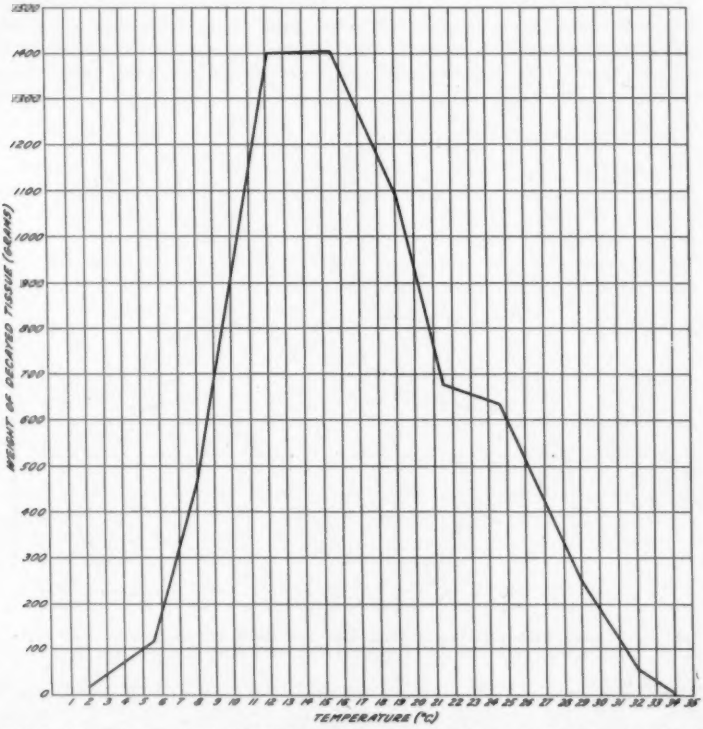

FIG. 5.—Decay (in grams), at different temperatures, of Yellow Jersey sweet potatoes inoculated with *Pythium ultimum*, after three days

The temperatures in the incubators did not vary more than 1 degree during the entire course of the experiment except in one incubator which dropped several degrees for a few hours as the result of a breakdown in the mechanical equipment. This mishap, however, did not seem to alter the results to any perceptible degree. The temperatures were read twice daily.

The temperatures employed and the amount of decay at each are shown in Figure 5. The results, especially at the optimum, are in sharp contrast to what might be expected if the growth in pure agar cultures is used as a guide. A little decay took place at 2° and 34° C., but as these temperatures are never maintained in a storage house, they are of little practical consequence. The amount of decay rose

sharply with the increase in temperature to 12° and slightly more to 15.5°. The optimum, therefore, probably lies somewhere between 12° and 15.5°. From 15.5° the amount of decay decreases with the rise in temperature and amounts to practically nothing at 34°. This organism, with an optimum for growth of about 32° in pure cultures on cornmeal agar, produces the optimum amount of decay at a temperature much lower (12° to 15.5°). These results suggest that it is unsafe to draw any general and sweeping conclusions with respect to the optimum temperatures for decay from growth in artificial culture media. Each will need to be determined separately.

From the results of these investigations, it is apparent that the minimum temperature for infection is approximately 2° C.; the optimum, 12° to 15.5°; and the maximum, 34° to 35°. The minimum, optimum, and maximum are practically the same for infection of the Triumph and Yellow Jersey varieties of sweet potatoes.

In the infection tests conducted in the laboratory at room temperature (22–25° C.), it was noted that the same symptoms were not always produced when parallel experiments were carried out with the same organism. This was true with both *Pythium ultimum* and *P. scleroteichum*, and at times both produced the marble and the cheesy type of decay, a phenomenon which upset the a priori conception of the causal relationship of these organisms to mottle necrosis. In further defense of the nonspecificity of these two organisms for different types of decay, the studies on the effect of temperature on the amount of decay produced show that there is a correlation between the temperature and the type of decay produced. At temperatures from 2° to about 18° C. a cheesy type of rot was produced and the color was almost identical with the healthy tissue. The cavity produced was more or less uniform in diameter, with relatively smooth walls. Above 18°, however, the decayed tissue was more or less crumbly and mealy, a condition which became more pronounced with the rise in temperature. At the higher temperatures cavities of irregular shape and sizes were produced and the walls were more or less ragged and shredded.

These experimental data have a direct bearing on what may be expected, and what actually does happen, in the field. Experience has shown that the type of decay is more or less correlated with the temperature of the soil. If the weather is warm, as is likely in late summer, the marble type will result. If, on the other hand, the weather is cool following infection, the cheesy type will predominate. It was shown by several series of inoculation experiments with the same organism that the higher the temperature the greater the tendency to marbling. There was, as would be expected, an intermediate temperature at which either one or both might occur. At the optimum temperature for decay the cheesy type predominates, indicating that the marbling, as found in the field, occurs as a result of abnormal environmental conditions. Inasmuch as the optimum temperature for decay is low, the absence of the disease in some seasons might be explained on the basis of the high temperatures sometimes prevailing in the summer and fall. Further evidence of this is the fact that most of the initial infections which usually occur in late summer or early autumn take place at the lower ends of the edible roots, which are embedded in the cooler rather than in the upper warmer strata of soil.

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STORAGE

While any sweet potatoes noticeably infected with mottle necrosis would be discarded in digging time, some are likely to be overlooked and therefore would find their way into storage. Not only has a careful study been made of this kind of material, but mottle-necrosis material has been collected and exposed to different temperatures and humidities. For example, diseased potatoes have been stored under normal conditions, where the temperatures and humidity would fluctuate to some extent. At digging time (October 12 to 15) infected sweet potatoes also have been exposed to a temperature of from 22° to 25° C., with a relative humidity of about 90, and in an incubator with a temperature varying from 9° to 10°. At frequent intervals thereafter some of the sweet potatoes were removed and cultures made from diseased tissue. In the early part of the storage period Pythium could be isolated with ease, but, as time went on, this became increasingly difficult. Some of these sweet potatoes were kept in storage until the following March, but Pythium was not isolated later than November 13. It would seem from these results that the organism does not live long in dormant roots under storage conditions. Numerous microscopic examinations of necrotic tissue revealed the presence of a Pythium in and about the cells, but it lacked the appearance of a living organism. As it became more difficult to isolate Pythium, other fungi—such as Fusarium and Trichoderma—were obtained with greater ease and frequency. These latter organisms had apparently invaded the tissue decayed by Pythium and, it is not improbable, may have destroyed it. Certain results seem to indicate that Pythium is relatively short-lived when grown on artificial media; also that it is very sensitive to toxins such as might be released by other fungi growing in competition with it. At lower temperatures (9–10°) Pythium was isolated up to November 6. After this date *Mucor racemosus* Fes. only was obtained. This organism frequently causes considerable damage to sweet potatoes at low temperatures when the relative humidity is fairly high.

Whether or not any additional decay in storage is caused by Pythium has not been definitely determined. It is believed that there is little or none. Although Pythium in pure culture will rot sweet potatoes with ease, it seems to succumb readily to the competition of other fungi. While mottle-necrosis sweet potatoes have been examined during the storage period, no data have been collected with respect to the increased amount of decay by Pythium under natural storage conditions. In fact, such data would be difficult to obtain because of the presence of other fungi and the difficulty of measuring the amount of decay at different times without destroying the specimen. There is reason to believe, however, that other fungi, Fusarium, for example, enter mottle-necrosis tissue and continue to decay the sweet potato. Potatoes affected with mottle necrosis after a few weeks in storage often reveal a condition scarcely typical of the disease. The chambers and armlike ramifications through the potato characteristic of mottle necrosis are often no longer present. In other words, the sweet potato frequently is uniformly decayed from one end or one side in a manner resembling end rot caused by a Fusarium, or, if the temperature is rather low, by *Mucor racemosus*, which causes symptoms closely resembling those produced by Pythium.

In general, the writers are strongly of the opinion that Pythium is not a storage-rot-producing organism, but that if sweet potatoes infected in the field are stored the decay probably is continued by other fungi.

CONTROL

From the present knowledge of mottle necrosis, some recommendations which should help to control the disease can safely be made. The prevalence and destructiveness of the disease depends more or less on certain environmental conditions such as soil moisture and temperature. The disease is worse in certain types of soils, particularly in the medium light ones. It has been shown that the rootlet rot, caused by the same organism as mottle necrosis, occurs in the hotbed and that it is increased by using the same soil year after year. In view of these facts, it is advisable to use new soil or sand each year in the hotbed. Pythium succeeds best in soil or sand containing a considerable amount of moisture. Care should be exercised, therefore, not to apply any more water to the plants in the hotbed than is actually necessary. During the several years that these investigations have been under way the same ground in the field has been used for two or three years in succession. The results of this practice seem to have actually increased Pythium in the soil. Of course there is no actual proof of this, as the prevalence of the disease in the field is dependent—partially at least—on weather conditions, which were probably not identical for the different years. Nevertheless, where this disease has been destructive, it is advisable to plant the ground to other crops.

Ever since investigations with this disease were undertaken attention has been given to the comparative susceptibility and resistance of the sweet-potato varieties to mottle necrosis. Results recorded above have shown that all the varieties can be infected artifically. On the other hand, these varieties have been planted on infected soil, with the result that a considerable degree of resistance on the part of some of them was shown under field conditions. The Triumph, Yellow Jersey, and Big Stem Jersey have proved to be the most susceptible, and a little mottle necrosis has been found on the Georgia and Gold Skin in the field, but for the most part the remaining varieties listed in Table 2 may be regarded as immune, at least for all practical purposes. There is always, therefore, the possibility of substituting some of the immune varieties when the susceptible ones can not be grown.

RHIZOCTONIA

While the purpose of this paper is to discuss a disease of sweet potatoes caused by Pythium, it seems desirable, in order to avoid confusion, to discuss a similar condition in the field caused by *Rhizoctonia solani* Kühn. Attention has already been called to the prevalence of Pythium on the small roots and rootlets in the field and in the hotbed. During the summer of 1925, and to a lesser extent in 1924 and 1926, the roots were badly diseased in the sandier portion of the field. So far as macroscopic appearance went, the disease somewhat resembled Pythium rootlet rot, and it was so diagnosed. However, as a result of a large number of isolations it was found that *R. solani* was quite as prevalent as Pythium. Attention is called to

the presence of Rhizoctonia in order to avoid confusion in the diagnosis of the disease in the field. When it was learned that both organisms were present, it was possible in some cases to separate the two. *R. solani* causes an almost black soft decay of the roots from the tip to the main stem. The cortex of the root is loosened and often can be separated easily from the xylem. Pythium, on the other hand, usually begins at the root tip. It attacks, as a rule, smaller roots than does Rhizoctonia. The death of many roots from attacks of Rhizoctonia often results in the development of a large cluster of small roots, roughly resembling a witches' broom (fig. 6).

Rhizoctonia solani, regardless of how prevalent it may be in the field, has not been found associated with mottle necrosis in any way.

SUMMARY

A field disease of sweet potatoes (*Ipomoea batatas* Poir.), known as mottle necrosis, has been described. The disease occurs very commonly on the Triumph, Yellow Jersey, and Big Stem Jersey, and, to a lesser extent, on some of the other common commercial varieties. When conditions are favorable for the disease as much as 40 to 50 per cent of the crop may be destroyed.

Several species of Pythium will produce mottle necrosis. Inoculation experiments have shown that it can be produced readily by *Pythium ultimum* Trow, *P. scleroteichum* Drechsler, and *P. aphanidermatum* (Edson) Fitzpatrick, and to a lesser extent by *P. splendens* Braun and *P.* 5825. *Pythium ultimum* and *P. scleroteichum* were isolated from rootlets in the hotbed. Therefore, the same organisms which cause rootlet rot cause mottle necrosis.

In the field three varieties of sweet potatoes (Triumph, Yellow Jersey, Big Stem Jersey) are very susceptible. All varieties tried were infected artificially.

The type of decay depends largely on the temperature. If the temperature is 20° C. or above, it is likely to be marbled; if lower, the cheesy type of decay will predominate.

The optimum for the growth of *Pythium ultimum* and *P. aphanidermatum* in pure culture on corn-meal agar is 32° C.; for *P. scleroteichum*, 29°. The minimum temperatures are approximately as follows: *P. ultimum*, slightly below 2°; *P. aphanidermatum*, about 6°; *P. scleroteichum*, about 8°. The maximum temperature for *P. ultimum* is approximately 41.8°, for *P. scleroteichum* about 38.5°, and *P. aphanidermatum* about 41.8°.

The results of these investigations show that, although the optimum temperature for growth of *Pythium ultimum* in pure culture is about 32° C., the greatest amount of decay of sweet potatoes is produced in a given time (five days) at temperatures between 12° and 15°. The optimum temperature for growth in pure culture and the optimum temperature for decay are quite dissimilar.

Mottle necrosis has not been found to be a storage trouble.

Rhizoctonia solani Kühn has been found to cause decay of some of the small roots of sweet potatoes in the field. The symptoms produced resemble somewhat rootlet rot caused by Pythium, but differ from the latter in that the roots are more completely destroyed, and also in that clusters of small roots subsequently develop which re-

semble a witches' broom. Rhizoctonia seems to be more prevalent on light sandy soils. It is in no way associated with mottle necrosis.

Crop rotation should be practiced where control measures are required. New soil should be used each year in the hotbed. While

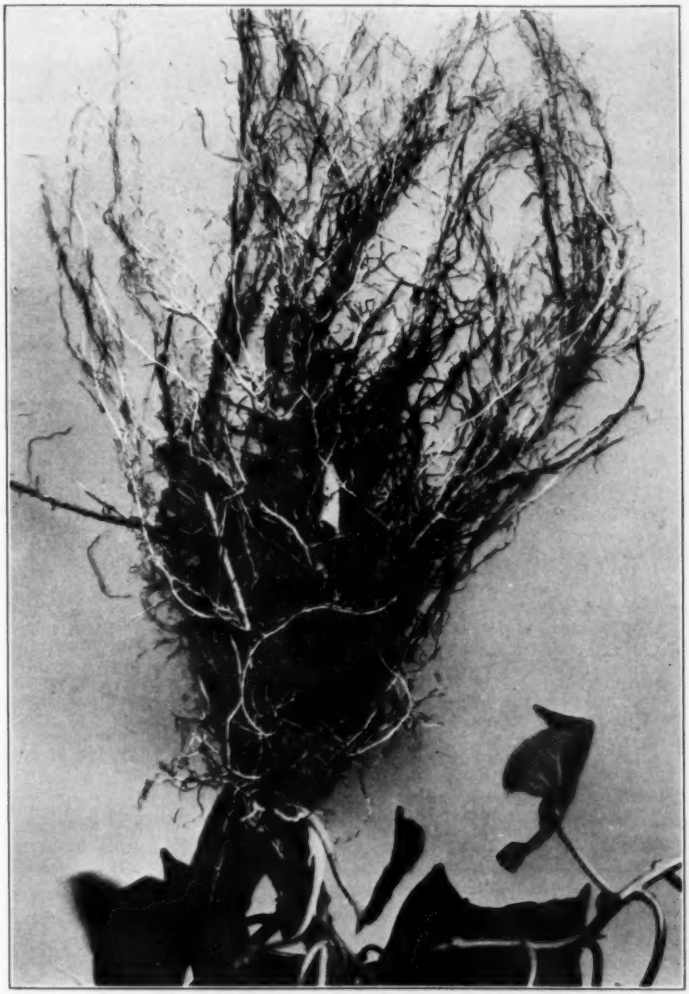

FIG. 6.—Excessive root development, resembling a witches' broom, on sweet potato, probably caused by *Rhizoctonia solani*. (About three-fifths natural size.)

all the commercial varieties have been infected artificially, only the Triumph, Yellow Jersey, and Big Stem Jersey have been found seriously diseased in the field. A number of good commercial varieties appear to be more or less immune.

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THE COMPARATIVE SUSCEPTIBILITY OF SWEET-POTATO VARIETIES TO STEM ROT [1]

By L. L. HARTER, *Pathologist*, and W. A. WHITNEY, *Junior Pathologist, Office of Vegetable and Forage Diseases, Bureau of Plant Industry, United States Department of Agriculture*

INTRODUCTION

Investigations [2] have shown that sweet potatoes (*Ipomoea batatas* Poir.) are affected with a disease known as stem rot or wilt (caused by *Fusarium batatatis* Wr. and *F. hyperoxysporum* Wr.), whose distribution throughout the United States is more or less correlated with the distribution of the susceptible varieties. In some regions of the South stem rot is unknown or causes little or no loss, owing to the fact that the varieties of sweet potatoes there grown are immune or highly resistant and suffer no apparent injury from it. On the other hand, in the northern tier of the sweet-potato-growing States localities may be found where stem rot is so severe on the small number of varieties grown—and practically all of them are very susceptible to the disease—that it threatens to destroy the industry or to force it out of its present centers.

Soil, climate, and commercial competition with other crops have confined the sweet potato in the northern range to relatively small areas in several States (New Jersey, Delaware, Maryland, Virginia, Ohio, Indiana, Illinois, Iowa, Kansas, and California) where a susceptible variety is generally grown, sometimes for 3 to 12 or 15 years on the same soil without rotation with other crops. Not only is this a bad agricultural practice but it causes the soil to become thoroughly infested with the causal organisms. All of the plants being potential hosts, many of them become actually infected.

In the South the situation is somewhat different. A large number of varieties are grown there, many of which are apparently resistant to stem rot or are not injured by it. If any particular variety is found to be damaged by the disease, a resistant one, probably equally acceptable to the consumer, can be substituted for it.

The disease situation has become so serious in some of the Northern States that a series of experiments were undertaken which had for their object to determine, first, the comparative susceptibility of a number of commercial varieties and, second, the adaptability of southern-grown varieties to northern conditions with the view of selecting varieties showing a suitable degree of resistance to stem rot. Not all the known varieties of sweet potatoes were tried in these experiments, the collection consisting of those of established economic

[1] Received for publication Mar. 12, 1927; issued July, 1927.

[2] HARTER, L. L., and FIELD, E. C. THE STEM-ROT OF THE SWEET POTATO (IPOMOEA BATATAS). Phytopathology 4: 279-304, illus. 1914.

importance and others of commercial promise. The following varieties were used: Big Stem Jersey, Creola, Dahomey, Dooley, General Grant Vineless, Georgia, Gold Skin, Haiti, Key West, Nancy Hall, Pierson, Porto Rico, Pumpkin, Red Brazil, Red Jersey, Southern Queen, Triumph, White Yam, Yellow Jersey, Yellow Strasburg, Yellow Yam.

In 1914 Harter and Field [3] studied the susceptibility of several varieties of sweet potatoes to stem rot by making artificial inoculations in the field. A few varieties were also planted on infested soil in New Jersey. The results of these several experiments showed that a number of the varieties could be infected artificially; also, that natural infection of a few varieties occurred when planted on infested soil. Inasmuch as plants often may be inoculated artificially when, under normal field conditions, they might remain healthy, it was planned to conduct a series of field tests, using, where practicable, only naturally infested soil.

METHODS OF EXPERIMENTS

A 1-acre plot was selected at Houston, Del., and the varieties were grown on this field for four consecutive years, beginning with 1922. Another plot of a similar area at Seaford, Del., where stem rot was very prevalent, was planted to the same varieties for two consecutive years, 1925 and 1926. Stem rot, though present, was not especially abundant in the Houston soil, and in order to increase the chances for infection and infestation of the soil the roots of the plants used in 1922 and 1923 were dipped in a spore suspension of the stem rot organisms just previous to planting. About 500 plants of each variety were set out at both Houston and Seaford. Counts of diseased and healthy plants were made about every 10 days to 2 weeks from the time of setting until the vines covered the field so that the individual hills could no longer be recognized. At each count the diseased plants were pulled up and removed from the field. In the fall when the crop was dug each individual plant was carefully examined for stem rot.

The original plan of this variety test called for a five-year planting on the 1-acre plot selected at Houston, Del. Because of the depredations caused by chickens and other barnyard animals, it was thought advisable to check the results by duplicating the experiments for two years at Seaford, Del. The latter plot was located in the center of a field where a large percentage of the plants were infected the previous year. The soil, ideally suited to the growing of sweet potatoes, was well prepared and the crops received the best of care throughout the growing season. Notwithstanding the fact that conditions on the Houston plot were not of the best, the results, when compared with those obtained at Seaford, are strikingly similar.

The seed potatoes of the different varieties, with two or three exceptions, were obtained from the Office of Horticulture, Bureau of Plant Industry, United States Department of Agriculture. The plants were grown at the Arlington Experiment Farm near Rosslyn, Va., and shipped by express to Houston and Seaford when required.

[3] HARTER, L. L., and FIELD (TILLOTSON), E. C. EXPERIMENTS ON THE SUSCEPTIBILITY OF SWEET POTATO VARIETIES TO STEM ROT. Phytopathology 5: 163–168. 1915.

EXPERIMENTAL DATA

Figure 1 shows in graphic form the reaction of different varieties of sweet potatoes to stem-rot infection at both Houston and Seaford, Del. The curves shown in Figure 1 were plotted from the combined

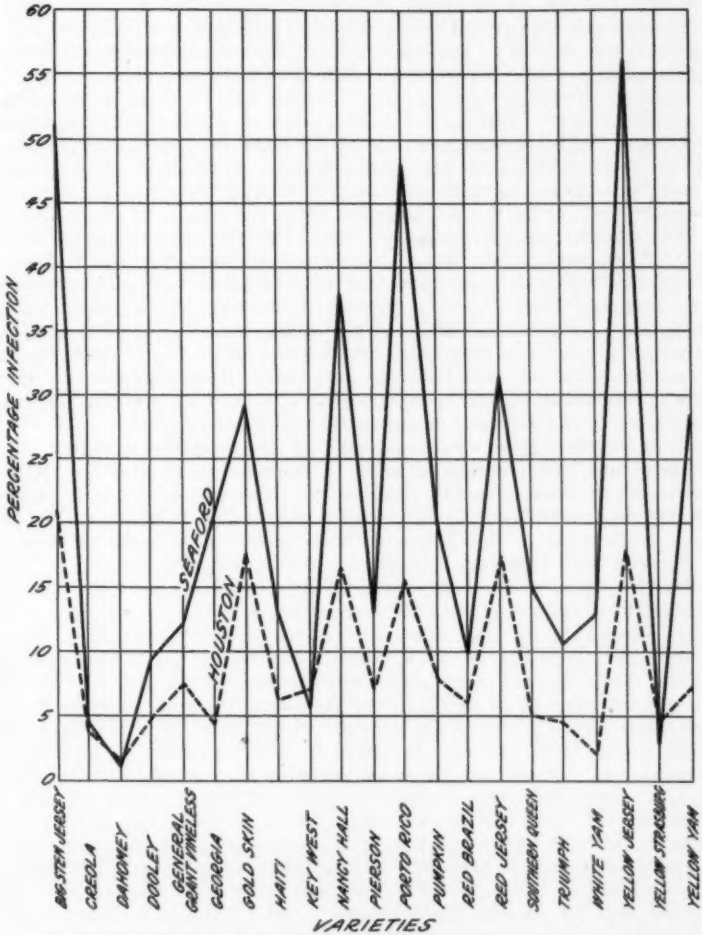

FIG. 1.—Percentage infection of different varieties of sweet potatoes by the stem-rot organisms for the period of experiment—four years at Houston and two years at Seaford, Del.

results of a four-year and a two-year test at Houston and Seaford, respectively. The total number of plants of each variety grown at each station and of those diseased (including both those removed during the season and at the final harvest count) was used as a basis for calculating the percentage of infections. As would be expected, there was some variation from year to year among the varieties, and

during seasons especially favorable to infection the percentage of diseased plants was noticeably higher.

A careful examination of these curves shows that there is, for the most part, a marked degree of similarity of infection of the same varieties grown at the two stations. One notable exception is the Georgia variety which shows 4.5 per cent infection at Houston and 27.5 per cent at Seaford. The results at Houston are surprising, in view of the fact that this variety has been found quite susceptible when observed in a number of different locations.

Special attention should be called to the results of two other varieties, White Yam and Haiti. Both of these varieties usually exhibit a high degree of resistance to stem rot, the percentages at Houston being 2.1 and 6.3, respectively. At Seaford, on the other hand, in a badly infested soil, the infection increased to 13.1 and 13, respectively, on the same varieties.

An examination of the curves shows that the percentage of infection of the same varieties is, in general, higher at Seaford than at Houston. Six varieties stand out as especially susceptible to stem rot, namely, Yellow Jersey, Big Stem Jersey, Porto Rico, Nancy Hall, Red Jersey, Gold Skin, and Georgia. Yellow Jersey and Big Stem Jersey, with percentages of infection of 56.3 and 51.1, respectively, at Seaford, are followed by Porto Rico and Nancy Hall. There is nothing surprising in these results, for the varieties are those that have long been recognized as the most susceptible.

It is evident, however, that none of the varieties tried in these experiments are entirely immune. Poole [4] found Red Brazil and Triumph to be practically immune to stem rot under New Jersey conditions, there being either no plants killed or infections were less than 1 per cent. The Triumph was found to be highly resistant. It showed only 2 per cent diseased plants during the season.

Judging from the results of the writers' investigations, none of the 21 varieties studied in these experiments are entirely immune to stem rot. Obviously cultures were not made from the thousands of plants examined. Whether or not a plant was infected was based entirely on the characteristic macroscopic appearances of the plants. During the several counts in the summer following planting, infection was determined mostly from the general appearances of the foliage and occasionally from the discoloration of the fibrovascular bundles at the soil line. When the diseased plants were counted and removed at this time only those unquestionably infected were classed as diseased, the doubtful ones being left for observation at the next count. All the varieties showed a certain percentage of infection at these early counts and some of the plants of more or less resistant varieties were killed. The percentage of killed plants, however, is considerably smaller than Figure 1, which includes those barely infected as well as those actually killed, indicates. The fact that a few plants of all varieties are actually killed is proof of the statement already made that none of the varieties are actually immune.

Just when a plant is or should be classed as infected is sometimes difficult to tell, and it is not unlikely that different observers well

[4] POOLE, R. F. THE STEM ROT OF SWEET POTATOES. LOSSES, SOURCES OF INFECTION AND CONTOL. N. J. Agr. Expt. Sta. Bul. 401, 32 p., illus. 1924.

——— CULTURAL METHODS FOR REDUCING SWEET POTATO LOSSES CAUSED BY STEM ROT. N. J. Agr. Expt. Sta. Bul. 433, 16 p., illus. 1926.

acquainted with the disease would not always agree. On the one hand there are the unmistakable symptoms such as death of the plant and the blackening of the fibrovascular bundles about which there can be no question, but on the other hand there are those doubtful cases which are on the border line whose classification rests wholly upon the judgment of the observer. At the time of digging there was a large number of such border-line cases especially among those varieties exhibiting on the whole a considerable degree of resistance (Dahomey, Triumph, Haiti, Red Brazil, White Yam, and others). When the plants were examined in the fall, the roots of each plant were cut open with a knife. In some cases it was found that the causal fungus had gained entrance at the wound made by pulling the plant from the mother potato and had extended for 1 or 2 inches up into the stem. It was also found that the stem-rot fungus would sometimes enter through a soil-rot (pox) lesion and invade the fibrovascular system for a short distance in either direction or that it would enter through other channels, such as wounds made in cultivation or by other means. It is interesting to note in this connection that there were no external symptoms of stem rot. The foliage was healthy, and the plants grew normally and produced a normal crop. A large percentage of the more or less resistant plants responded in this way, and in those cases where a plant was actually killed the infection took place in the spring soon after the plants were set out and before they had become established. There is, then, a number of varieties of this type which, while not entirely immune, are resistant enough to yield a normal crop on badly infested soil. The varieties that could be safely planted on infested soil are as follows: Creola, Dahomey, Haiti, Key West, Pierson, Pumpkin, Red Brazil, Southern Queen, Triumph, White Yam, and Yellow Strasburg.

SUMMARY

The relative susceptibility of 21 varieties of sweet potatoes to stem rot (caused by *Fusarium batatatis* Wr. and *F. hyperoxysporum* Wr.) was tested four years at Houston, Del., and two years at Seaford, Del., on naturally infested soil. At Houston the infestation of the soil and the chance for infection were increased by dipping the roots of the plants in a spore suspension of the stem-rot organisms just previous to planting.

The two curves (fig. 1), which are more or less strikingly parallel, show that those varieties very susceptible at Seaford were also very susceptible at Houston.

Judging from the results of these experiments, there are no varieties entirely immune to stem rot, but such varieties as Creola, Dahomey, Haiti, Key West, Pierson, Pumpkin, Red Brazil, Southern Queen, Triumph, White Yam, and Yellow Strasburg are so slightly injured by the invasion of the parasites that a normal crop can be produced by them even when they are grown on badly infested soil.

The following varieties are very susceptible to the disease: Yellow Jersey, Red Jersey, Porto Rico, Nancy Hall, Gold Skin, Georgia, and Big Stem Jersey. A large percentage of the infected plants of these varieties are killed during the growing season.

INHERITANCE OF RATE OF SHEDDING IN A COTTON HYBRID[1]

By THOMAS H. KEARNEY, *Senior Physiologist in Charge*, and ROBERT H. PEEBLES, *Senior Scientific Aid, Office of Alkali and Drought Resistant Crops, Bureau of Plant Industry, United States Department of Agriculture*

INTRODUCTION

The tendency of the cotton plant to shed many of its flower buds and young bolls is so important in its relation to cotton production that no aspect of this phenomenon should be disregarded. Variations in the rate of shedding within a variety doubtless are determined chiefly by environmental factors, including such external biological factors as insects and diseases. But when different types of cotton are compared as to their shedding percentages, the probability that genetic factors also are involved must not be overlooked.

The writers have presented evidence that different species and varieties of cotton differ consistently in the rate of shedding. They have shown also that in a hybrid between Pima Egyptian and Acala upland cottons the second generation was more variable than the first and that individual F_2 plants grown under identical conditions differed significantly in the percentage of buds and of young bolls lost by abscission.[2] Third-generation progenies have been grown subsequently, and the data obtained from them are set forth in the present paper. The new evidence confirms the conclusion that abscission of the flower buds and young bolls in cotton is determined partly by genetic factors.

COMPARISON OF F_3 PROGENIES WITH THEIR F_2 PARENTS

The investigation was conducted at the United States Field Station, Sacaton, Ariz. Fifteen individuals of the second generation of the hybrid Pima × Acala were selected in 1925, of which five had shown a high rate of shedding, five an intermediate rate of shedding, and five a low rate of shedding. Third-generation progenies were grown in 1926 from self-pollinated seed of the F_2 plants, and a record was kept for each plant of the total number of flower buds and flowers produced and of the losses by abscission, both before and after anthesis. The method of taking the shedding records is described by the writers in a previous paper.[3] From the resulting data were computed the shedding percentages of the individual plants and the mean and standard deviation of the mean of each F_3 progeny.

[1] Received for publication Mar. 12, 1927; issued July, 1927.

[2] KEARNEY, T. H., and PEEBLES, R. H. HERITABILITY OF DIFFERENT RATES OF SHEDDING IN COTTON. Jour. Agr. Research 33: 651-661, illus. 1926.

[3] KEARNEY, T. H., and PEEBLES, R. H. Op. cit. p. 653.

The mean shedding percentages of each progeny were computed by taking, for all plants as one array, the number of flower buds shed, the number of young bolls shed, and the number of buds shed plus the number of bolls shed as percentages of the total number of flower buds produced; also the number of bolls shed as a percentage of the number of flowers (buds which reached the stage of anthesis). The standard deviations of these percentages were computed with regard to variation among the individual plants of the progeny by the formula

$$\sigma = \sqrt{\frac{\Sigma n p^2}{\Sigma n} - Mp^2}$$

where p = the shedding percentage of each plant, n = the number of buds or of flowers produced on each plant, and Mp = the mean shedding percentage of the progeny. The standard deviations computed by this formula were then increased to take account of the small numbers of plants in these populations, and the probable errors were computed from the standard deviations as thus corrected.[4] The standard deviations as given in Tables 1 and 2 have been further increased to bring them to the values expected had the ratio been 0.5 or 50 per cent,[5] thus making the standard deviations of large and of small percentages comparable as expressions of the variability of the several populations. This second correction was not used in computing the probable errors as here given, although it was erroneously so used in the paper cited.

The shedding percentages of the F_2 parents and the statistical constants of the F_3 progenies are given in Table 1, in which all percentages are based upon the total number of flower buds produced by the F_2 plant or the F_3 progeny; and in Table 2, in which the boll-shedding percentage is based upon the total number of flowers produced by the F_2 plant or the F_3 progeny.[6] The progenies are arranged in both tables in the order of the total shedding percentages of the F_2 parents.

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[4] In computing the probable error by the formula $E = .6745 \frac{\sigma}{\sqrt{n}}$, n was taken as the number of plants in the progeny, and not as the number of buds or of flowers.

[5] KEARNEY, T. H., and PEEBLES, R. H. Op. cit. p. 654, footnote.

[6] On seven of the F_2 plants the time of shedding, whether before or after anthesis, was not recorded for some of the "sheds," the number unclassified having constituted from 5 to 33 per cent of the total number of buds and bolls shed by these plants. The unclassified "sheds" were arbitrarily divided between buds and bolls in the proportion indicated by the known numbers of buds and bolls shed by the plant in question. The bud-shedding and boll-shedding percentages of these plants are correspondingly uncertain, but the departure from the true percentage could in no case be large enough to affect the conclusions. The total shedding percentage is not affected by this uncertainty of classification.

TABLE 1.—*Shedding percentages of 15 F_2 plants and their F_3 progenies of a hybrid between Pima Egyptian and Acala upland cottons, the number of buds and of bolls shed being taken as a percentage of the number of flower buds produced*

Designation of F_2 parent and F_3 progeny	Number of plants in F_3 progeny	Percentage shedding before anthesis (bud shedding)			Percentage shedding after anthesis (boll shedding)			Percentage total shedding (bud and boll shedding)		
		F_2 individual	F_3 progeny		F_2 individual	F_3 progeny		F_2 individual	F_3 progeny	
			Mean	Standard deviation		Mean	Standard deviation		Mean	Standard deviation
High group:										
4-15-24	18	47.5	58.2±4.5	28.8	42.5	23.4±2.6	19.5	90.0	81.6±2.4	19.5
7-17-13	6	49.2	40.3±3.5	13.0	38.8	48.3±4.4	15.9	88.0	88.6±1.0	5.7
18-2-20	8	19.2	24.7±2.0	9.7	68.3	54.3±2.4	9.9	87.5	79.0±1.5	7.6
17-13-20	11	15.1	58.2±4.4	21.7	67.7	27.2±3.6	20.1	82.8	85.4±2.1	10.6
4-1-22	16	27.6	45.5±3.5	20.7	53.9	38.2±2.8	17.1	81.5	83.7±1.7	13.6
Average		31.7±5.1	45.4±4.5		54.2±4.4	38.3±4.2		86.0±1.2	83.7±1.2	
Intermediate group:										
4-15-6	16	22.0	24.9±3.0	20.5	40.6	33.5±2.1	13.0	62.6	58.4±2.3	13.9
4-15-14	8	31.0	56.2±3.4	14.6	31.0	25.0±2.2	10.7	62.0	81.2±2.1	11.5
7-1-21	12	32.3	22.5±2.9	17.8	28.2	48.6±4.7	24.2	60.5	71.1±4.1	22.9
17-13-9	11	13.5	49.1±3.8	18.8	45.8	17.2±1.9	12.6	59.3	66.3±3.0	15.4
7-1-22	18	15.9	46.6±4.8	30.3	42.0	35.2±3.7	24.4	57.9	81.8±1.5	12.4
Average		22.9±2.7	39.9±4.9		37.5±2.4	31.9±3.8		60.5±0.6	71.8±3.2	
Low group:										
17-16-12	15	10.8	28.0±3.3	21.3	29.4	27.9±2.3	14.6	40.2	55.9±3.1	17.7
7-1-1	14	19.8	56.7±2.8	15.8	19.8	10.1±1.3	11.6	39.6	66.8±2.2	12.7
17-13-3	18	6.0	29.9±2.8	19.2	29.8	23.1±1.9	14.3	35.8	53.0±2.2	13.8
17-13-12	13	8.7	29.2±4.8	28.4	26.3	45.3±3.7	20.0	35.0	74.5±1.7	10.5
18-7-10	11	3.9	34.0±4.0	20.8	18.0	29.1±3.6	19.5	21.9	63.1±4.4	22.5
Average		9.8±2.0	35.6±3.9		24.7±1.8	27.1±4.1		34.5±2.4	62.7±2.8	

TABLE 2.—*Boll-shedding percentages of 15 F_2 plants and their F_3 progenies, of a hybrid between Pima Egyptian and Acala upland cottons, the number of bolls shed (abscissions after anthesis) being taken as a percentage of the number of flower buds which reached anthesis*

Designation of F_2 parent and F_3 progeny	Boll-shedding percentage		Standard deviation of F_3 mean
	F_2 individual	F_3 progeny mean	
High group:			
4-15-24	81.2	56.0±3.6	17.8
7-17-13	76.6	80.9±3.8	17.7
18-2-20	84.5	72.2±1.9	8.9
17-13-20	79.8	65.0±3.8	19.4
4-1-22	74.6	70.1±2.7	17.3
Average	79.3±1.1	68.8±2.9	
Intermediate group:			
4-15-6	52.1	44.6±2.1	12.3
4-15-14	44.9	57.1±4.6	15.5
7-1-21	41.6	62.7±5.1	27.3
17-13-9	53.0	33.8±2.9	15.3
7-1-22	50.0	65.9±2.0	13.0
Average	48.3±1.6	52.8±4.3	
Low group:			
17-16-12	32.9	38.7±2.9	17.1
7-1-1	24.7	23.3±2.3	15.3
17-13-3	31.7	33.0±2.6	17.1
17-13-12	28.8	64.0±2.0	11.2
18-7-10	18.7	44.1±5.0	24.9
Average	27.4±1.9	40.6±4.9	

The data given in Tables 1 and 2 show considerable variation within each group (high, intermediate, and low) both as regards the means of the F_3 progenies and the standard deviations of the means. When the groups are compared on the basis of averages,[7] however, a consistent relation is observed. The group of high-shedding F_2 plants gave the highest average in F_3 for bud-shedding percentage, boll-shedding percentage, and total shedding percentage, all based on total buds (Table 1) and for boll-shedding percentage based on total flowers (Table 2). Similarly, the group of low-shedding F_2 plants gave the lowest average in F_3 for all shedding percentages and the group of intermediate F_2's gave intermediate averages in F_3. The differences among the averages of the three groups of F_3 progenies are greatest in respect to total shedding based on total buds (Table 1) and boll shedding based on total flowers (Table 2). The difference in F_3 between the average of the high and the average of the low group was seven times its probable error for total shedding percentage (Table 1) and five times its probable error for boll-shedding percentage (Table 2).

There was little correspondence between the individual F_2 plants and their F_3 progenies in bud-shedding percentage and boll-shedding percentage based on total buds (Table 1), the coefficient of parent-offspring correlation having been, for the bud-shedding percentage 0.289 ± 0.160, and for the boll-shedding percentage as thus computed 0.272 ± 0.161. On the other hand, the coefficient of parent-offspring correlation (F_2 with F_3) for the total shedding percentage was high and significant, being 0.715 ± 0.085, or more than eight times its probable error. There was also a fairly high and significant parent-offspring correlation for boll-shedding percentage based on total flowers (Table 2) r being 0.621 ± 0.107.[8]

The data from F_3 therefore support the conclusion suggested by study of the F_2 population in 1925 that shedding is partly controlled by genetic factors which, following hybridization, segregate and recombine, although no definite Mendelian ratio is observable.

BUD SHEDDING IN RELATION TO BOLL SHEDDING

Further evidence of the incidence of genetic factors in shedding was sought by examining the relation between abscission before and after anthesis in the same populations. When the "sheds" occurring in both stages are taken as percentages of the total flower buds produced (Table 1), there was no significant correlation between the bud-shedding and boll-shedding percentages of the F_2 plants (r 0.166 ± 0.169), but the F_3 progeny means showed a rather high and very significant negative correlation ($r - 0.643 \pm 0.102$). This was to be expected in view of the fact that in F_3, in which the total shedding was in most cases heavier than in F_2, all of the progenies lost by abscission before and after anthesis (total shedding) more than 50 per cent of their flower buds. If most of the buds are shed before anthesis, obviously few will be left to shed after anthesis; and, conversely, heavy losses after the buds have reached anthesis implies

[7] The probable errors of these averages were computed from the departures of the several progeny means from the average for the group and were weighted for the small number, five, of progenies in each group.

[8] It is likely that these parent-offspring correlations are higher than would have been obtained if an F_3 progeny had been grown from each of the 155 plants in the F_2 population of 1925. Fifteen plants, of which five represented each rate of shedding (high, intermediate, and low) could hardly have constituted a fair sample of the F_2 population, since plants having an intermediate rate of shedding were much more numerous than the extremes. The coefficients are given merely as convenient expressions of the relations shown by comparison of the averages of the several groups.

that comparatively few could have been lost before anthesis. The relation seems to be a purely mechanical one, although, as was pointed out in the earlier paper,[9] physiological factors also may be involved if, as is often assumed, the cotton plant normally produces more flower buds than it can develop into mature bolls. In this case, if the greater part of the "self-pruning" is accomplished before anthesis, less will be necessary after anthesis, and vice versa.

The influence of these mechanical or physiological relations is eliminated when the bud-shedding percentage (Table 1) is correlated with the boll-shedding percentage based upon the number of flowers instead of the number of flower buds (Table 2). When this was done, a positive correlation was found in both generations, the coefficients having been 0.613 ± 0.109 in F_2 and 0.346 ± 0.153 in F_3. The F_3 coefficient is only 2.3 times its probable error, but since it is positive like the certainly significant F_2 coefficient (r/E 5.6) there can be little doubt that there is a tendency, in both generations, for plants which shed many of their flower buds to shed also many of their young bolls, and vice versa. The occurrence of this positive correlation may be taken as additional evidence that shedding in these hybrid populations is determined partly by genetic factors.

SHEDDING RATE HEAVIER IN F_3 THAN IN F_2

By comparing the averages of the three groups for total-shedding percentages (Table 1) it is evident that the agreement between F_2 and F_3 is much closer in the high group than in the intermediate and low groups. In the high group the F_3 average is slightly lower than the F_2 average, while in the intermediate group F_3 gives a considerably higher and in the low group a much higher average percentage of total shedding than F_2. Similar relations are shown by the boll-shedding percentages based on total flowers (Table 2). In the high group the average for F_3 is somewhat lower than for F_2, while in the intermediate group F_3 gives a slightly higher average and in the low group a much higher average than F_2.

Shedding was so nearly complete in the high group of F_2 plants that their F_3 progenies could not be expected to show increases of the same magnitude as in the intermediate and low groups. But the absence of any increase at all in the averages for the high group is difficult to account for. An increased rate of shedding, if common to all three groups, would not have been surprising in view of the evidence from a similar interspecific cotton hybrid[10] that the average number of bolls retained per plant was much smaller for the F_3 progenies than for the corresponding F_2 parents.

Another explanation of the higher rate of shedding in F_3 than in F_2 would be afforded if it could be shown that seasonal conditions were more conducive to a high rate of shedding in 1926, when the F_2's were grown, than in 1925, when the F_2's were grown. If so, the difference should have been reflected in the rates of shedding of the parental varieties, Pima (Egyptian) and Acala (upland). No record of bud shedding of these varieties was made at Sacaton in 1926, but boll shedding was recorded on 100 plants of each variety by Harold F. Loomis of the Office of Cotton, Rubber, and Other Tropical Plants,

[9] KEARNEY, T. H., and PEEBLES, R. H. Op. cit. p. 658.
[10] KEARNEY, T. H. SEGREGATION AND CORRELATION OF CHARACTERS IN AN UPLAND-EGYPTIAN COTTON HYBRID. U. S. Dept. Agr. Bul. 1164, p. 42. 1923.

Bureau of Plant Industry. The resulting percentages, in comparison with those obtained by Mr. Loomis in 1925, are given in Table 3.[11] No significant difference between the two seasons is indicated by these data, and the only explanation remaining for the heavier shedding of the hybrid population in 1926 is that the tendency to sterility in this interspecific cotton hybrid is more pronounced in F_3 than in F_2.

TABLE 3.—*Boll-shedding percentages of the parental varieties, Pima (Egyptian) and Acala (upland)* [a]

Year	Pima variety			Acala variety		
	Number of—		Boll-shedding percentage	Number of—		Boll-shedding percentage
	Plants	Flowers		Plants	Flowers	
1925	20	1,004	11.2±3.62	20	945	66.7±1.83
1926	100	3,962	9.9±0.40	100	7,008	68.8±0.68
Difference			1.3±3.64			2.1±1.95

[a] The percentages were computed (as in Table 2) from the total number of flower buds reaching anthesis and the total number of "sheds" occurring after anthesis in each population. The probable errors of the percentages are computed from the squared departures from the mean of the population of the shedding percentages of the individual plants, each weighted by the number of flowers on the plant.

Comparison of the boll-shedding percentages of the hybrids (Table 2) with those of populations representing the parental varieties (Table 3) shows that for the high group the average in F_2 was higher than that of Acala grown in the same year (1925), while the average in F_3 was the same as that of the Acala population of 1926. For the low group the average boll-shedding percentage in F_2 was more than twice that of Pima grown in 1925, and the average in F_3 was four times that of Pima grown in 1926. None of the 155 plants of the F_2 population of 1925 had a boll-shedding percentage as low as the average of the Pima population of that year.

CONCLUSIONS

Study of the second generation of the interspecific cotton hybrid Pima Egyptian × Acala upland suggested that the shedding of flower buds and young bolls is determined partly by genetic factors. Conclusive evidence that such is the case was afforded by third generation progenies of F_2 plants which had shown, respectively, a high, intermediate, and low rate of shedding. The coefficient of parent-offspring correlation (F_2 with F_3) was 0.715 ± 0.085 for the total-shedding percentage (buds and bolls shed as a percentage of total buds) and 0.621 ± 0.107 for the boll-shedding percentage (bolls shed as a percentage of total flowers).

Further evidence that genetic factors are involved is given by the positive correlation between rate of bud shedding and rate of boll shedding in both F_2 and F_3 of the hybrid. This fact proves that the inherited tendency to a high or a low rate of shedding expresses itself in abscission both before and after anthesis.

There was a tendency to greater sterility in F_3 than in F_2, as was noted also in an earlier investigation of an upland-Egyptian cotton hybrid, but the data at hand do not suggest a satisfactory genetic explanation of this difference between the two generations.

[11] The data in Table 3 accord with the results of previous years in showing a fairly consistent difference between Pima and Acala cottons in their rates of boll shedding at Sacaton. The ratio of the boll-shedding percentage of Pima to that of Acala was 1 to 4.5 in 1923, 1 to 4.2 in 1924, 1 to 6 in 1925, and 1 to 6.9 in 1926.

THE BASAL METABOLISM OF MATURE CHICKENS AND THE NET-ENERGY VALUE OF CORN[1]

By H. H. MITCHELL and W. T. HAINES, *Illinois Agricultural Experiment Station*

INTRODUCTION

One of the great contributions of Armsby to the science of animal nutrition was the demonstration that the requirements of animals for nutrients, and the values of feeds in covering these requirements, are two separate and distinct problems, requiring different experimental methods for their solution. The requirement of an animal for energy, for example, must involve a study of the heat production of the animal and of the energy content of animal tissues and secretions produced during growth, fattening, egg production, gestation, and lactation. The energy requirements are, therefore, best expressed in terms referring to the animal rather than to the feed; hence the measurement of animal requirements in terms of feed or digestible nutrients is an incomplete and an inaccurate measurement, however valuable it may be in practical animal husbandry.

According to Armsby (*1*),[2] the measurement of the true value of feeds as sources of energy to animals involves a determination of the energy content of the feed and of all losses of energy incurred in its utilization by the animal. If the latter are subtracted from the former, the "net-energy" value of the feed is obtained. A more complete definition of the actual energy value of a feed to the animal has never been proposed, nor one of greater significance as a guide to experimental investigation. It would seem that the same conception might be applied to other nutrients.

WORK OF OTHER INVESTIGATORS

While considerable work has been done upon the net-energy value of feeds for steers, practically no work of this character has been done with reference to other farm animals. The present investigation was undertaken as an initial study of the utilization of feed energy by chickens.

Gerhartz in 1914 reported (*2*) an extensive investigation on two hens concerned with the energy required in egg production. As one phase of the investigation some consideration was given to the question of the "work of digestion" of food, i. e., the loss of food energy as heat during digestion and assimilation. Upon comparing the increase in heat production of hens during a day in which food was consumed over that during a day of fast, with the intake of nutrients, Gerhartz concluded that Zuntz's factors for the estimation of the "work of digestion" could be applied to chickens.[3] However, the conclusion appears to be very poorly supported by the experimental data compared, since on the days of fast the heating effect of the

[1] Received for publication Feb. 3, 1927; issued July, 1927.

[2] Reference is made by number (italic) to "Literature cited," p. 943.

[3] Zuntz's factors (*2, p. 189*) obtained upon mammals are: 0.8 calorie per gram of protein consumed, 0.24 calorie per gram of fat, and 0.4 calorie per gram of carbohydrate.

meal consumed on the preceding evening can not be regarded as having been entirely dissipated in 10 to 20 hours, while on the days of feeding the full heating effect of the food can not be assumed to have been obtained in an experimental period of less than 12 hours' duration. In one of the feeding periods the hen consumed an egg laid only three hours before the termination of the respiration period; nevertheless, the full heating effect of the contained nutriment is assumed to have been obtained. In view of these facts, the close agreement that Gerhartz obtained in two comparisons between the observed increases in heat production and those computed by means of the Zuntz factors applied to the nutrients consumed must be considered purely fortuitous.

Hári (*4*) and Hári and Kriwuscha (*5*) have made observations on the heating effect of corn upon ducks and geese. Using the values obtained by direct calorimetry, the results may be summarized as shown in Table 1.

TABLE 1.—*The heating effect of corn on ducks and geese, as summarized from the experiments of Hári, and of Hári and Kriwuscha*

Bird	Body weight	Corn consumed	Heat production per day	Estimated basal heat production	Heating effect of corn	
					Observed	Calculated to 100 grams of corn
	Grams	*Grams*	*Calories*	*Calories*	*Calories*	*Calories*
Goose A	3,541	50	196	175	21	42
	3,499	100	274	172	102	102
	2,505	100	225	138	87	87
Goose B	3,126	50	241	153	88	176
	3,126	100	233	152	81	81
Duck A	967	50	127	90	37	74
Duck B	860	50	111	69	42	84

It is evident that the results obtained, calculated to 100 grams of corn, exhibited a rather wide variability, precluding any satisfactory estimate of a representative average.

In the same investigations the gross energy of food and solid and liquid excreta was determined, permitting an estimation of the metabolizable energy of corn for geese and ducks. The data covering metabolizable energy are summarized in Table 2.

TABLE 2.—*Metabolizable energy of corn for geese and ducks, as summarized from the experiments of Hári, and of Hári and Kriwuscha*

Bird	Body weight	Corn consumed	Gross energy of corn	Gross energy of excreta	Metabolizable energy of corn in—	
					Total calories	Percentage of gross energy
	Grams	*Grams*	*Calories*	*Calories*		
Goose A	3,541	50	202	38	164	81
	3,499	100	403	68	335	83
	2,505	100	403	80	323	80
Goose B	3,126	50	202	38	164	81
	3,126	100	403	65	338	84
Duck A	967	50	195	78	117	60
Duck B	860	50	195	75	120	62

For geese 82 per cent of the gross energy of corn appears to be metabolizable, while for ducks the much lower average of 61 per cent was obtained on two birds.

THE METABOLIZABLE ENERGY IN CORN

The first step in the present investigation was the determination of the metabolizable energy of corn. Results were obtained for 10 hens, 8 of which were Rhode Island Reds and 2 were White Plymouth Rocks. After several days' feeding of a constant amount of ground corn, the preliminary period being generally more than 10 days in length, the hens were placed in small cages, wire-mesh floors and movable pans being placed underneath for the collection of the excreta. The collection periods were seven days in length. The complete data resulting from this test are given in Table 3.

TABLE 3.—*Metabolizable energy of corn for hens*

Bird No.[a]	Average body weight	Average daily food	Daily energy intake	Energy of excreta per day	Metabolizable energy		
					Per day	Per 100 grams of corn	In terms of gross energy
	Grams	*Grams*	*Calories*	*Calories*	*Calories*	*Calories*	*Per cent*
129	2,126	44	181	28	153	348	84
130	1,610	44	181	31	150	341	83
132	1,407	42	172	30	142	338	83
135	1,732	44	181	31	150	341	83
74	2,144	50	196	32	164	328	84
129a	2,269	50	196	34	162	324	83
85	2,243	75	294	55	239	319	81
151	2,531	75	294	50	244	325	83
1		50	201	35	166	332	83
2	2,810	50	203	43	160	320	79

[a] Birds Nos. 1 and 2 were White Plymouth Rocks; the others were Rhode Island Reds.

The metabolizable energy of corn is taken as the difference between its gross-energy content and the gross energy contained in the excreta resulting from its digestion and assimilation. No correction has been made for the nitrogen balances of the birds. Such a correction is always small and can not be made with any great degree of accuracy, particularly with birds. It is also assumed that no appreciable amount of combustible gases is produced in the digestive tract of the chicken. The gross energy of the corn and excreta was determined in the Parr oxygen bomb calorimeter.

The percentage of the gross energy of corn that was metabolizable proved to be very constant, averaging 83.

All of the samples of corn used in these experiments were, unfortunately, not analyzed. However, that used for hens Nos. 74, 129a, 85, and 151 was found to contain 89.5 per cent dry matter, 3.49 per cent fat, and 1.56 per cent nitrogen, and to possess a gross-energy content of 3.917 calories per gram. The small variation in the composition of the samples of corn obtained for experimental work in this laboratory could hardly be considered as exerting any influence upon the percentage of its gross energy that would be metabolizable.

THE BASAL METABOLISM OF MATURE CHICKENS

Having determined the fraction of the gross energy of corn that is metabolizable, the next step in the investigation was to determine the basal heat production of mature chickens and the increase in heat production due to the consumption of corn. This heat increment represents a fraction of the metabolizable energy of corn that is inevitably wasted as heat. It is, therefore, just as surely an energy loss as the energy in the indigestible portion of the corn contained in the feces or that in the unoxidizable portion contained in the urine. Its determination is essential in arriving at a value for the net-energy content of corn.

The determination of heat production evidently requires the use of an animal calorimeter or respiration apparatus. In all of their calorimetric work on chickens the writers have used the gravimetric respiration apparatus of Haldane (*3*), as described and pictured in a previous publication from this laboratory (*10*). The writers have found that the method is simple of operation when its requirements have been carefully studied, economical of time, readily checked for possible errors, and capable of yielding consistent and accurate results except possibly for short observational periods. In the present investigation the observational periods were generally 22 to 23 hours in length. The weighing of the bottles and the feeding and watering of the chickens occupied the remaining one or two hours of the day. The rate of ventilation was approximately two liters per minute.

The heat production has been computed on the assumption that the total respiratory quotient is nonprotein. This method of calculation does not consider protein metabolism. However, from the data of Zuntz and Schumburg (*7, p. 62; 12*), relating to the calorific value of a liter of oxygen used in the combustion of the different nutrients, it may be computed that the maximum error in the total heat production figures resulting from this simplification is 6.58 per cent, and that the usual error would be less than 2 per cent.[4]

Total respiratory quotient	Maximum percentage of protein metabolism possible	Calorific value of a liter of O_2	Maximum error in assuming 100 per cent non-protein metabolism
0.75	44.65	4.594	3.06
.80	100.00	4.485	6.58
.85	73.12	4.624	4.92
.90	47.30	4.765	3.23
.95	22.97	4.906	1.58

Thus, if the total respiratory quotient is 0.85 and the protein metabolism is at its maximum (73.12 per cent of total calories), the error in neglecting the protein metabolism entirely would be less than 5 per cent. Under the much more usual conditions where the protein metabolism never exceeds 25 per cent of the total the error for all respiration quotients would be less than 1.5 per cent.

In the use of this or any other calorimetric method the temperature in the animal chamber must be under observation and should preferably be under control. During the daytime this could be

[4] Taking the calorific values of a liter of oxygen used in the combustion of protein, fat, and carbohydrate to be 4.485 calories, 4.686 calories, and 5.047 calories, respectively, the following is true:

accomplished readily. In view of the fact that the average critical temperature for Rhode Island Red hens is 62° (*10*), particular care was taken in this study that the temperature of the laboratory did not fall below 70° F. At night, however, control was not so effective, and occasionally the temperature fell considerably below 70°. A maximum and minimum thermometer was kept in the animal chamber in such position that its glass parts did not come in contact with the bird. The records read from this thermometer have been reported with all experiments.

The muscular activity of the chickens was also under continuous observation. The respiration chamber was suspended from a spring of such size that any considerable movement of the bird would result in an up-and-down movement of the chamber. This was recorded by a homemade "work adder" which revolved in only one direction. The record was, of course, purely of relative significance and served to assure the comparability of experimental periods as far as concerned the activity of the birds under observation. The animal chamber was not sufficiently large to permit of much movement, and the absolute darkness prevailing inside also tended to discourage much activity in the chicken. The work records have not been reported since (1) they indicate in general no great disproportion in activity among comparable experimental periods, (2) in the few cases in which a disproportionately large record was obtained the heat production of the bird was not out of line with that of comparable periods, and (3) the record has no definite or absolute significance with regard to the amount of movement within the chamber.

At the beginning it was considered necessary to determine how soon after feeding the heat production of the chicken returns to the basal level. From a large number of observations it was evident that, although the basal level was often reached after a fasting period of 24 hours, a fast of 48 hours was required before the basal level was reached in all cases. As illustrative of this finding, the data given in Table 4 are presented.

TABLE 4.—*Heat production of hens following the feeding of 75 grams of corn*

	Hen 2353				Hen 2001				Hen 200			
Day	Weight	Minimum and maximum temperature of chamber	Respiratory quotient	Heat production	Weight	Minimum and maximum temperature of chamber	Respiratory quotient	Heat production	Weight	Minimum and maximum temperature of chamber	Respiratory quotient	Heat production
	Grams			*Calories*	*Grams*			*Calories*	*Grams*			*Calories*
1	2,200	90–91	0.89	212	3,667	83–90	0.92	195	2,530	85–88	0.91	166
2	2,133	81–84	.94	153	3,533	82–90	.80	156	2,385	82–87	.79	[a] 144
3	2,047	85–90	.70	149	3,505	75–80	.69	151	2,287	80–93	.71	115
4	1,942	85–94	.72	[a] 154	3,463	70–86	.69	154	2,262	80–86	.71	110
5	1,897	89–95	.74	[b] 151	3,415	85–89	.70	134	2,230	77–87	.70	106

[a] Hen laid an egg weighing 52 gm. in respiration chamber.
[b] Eggshell found in respiration chamber. Apparently the hen had eaten contents.

TABLE 5.—*Basal heat production of mature chickens*

Bird No.	Date	Body weight	Body surface	Minimum and maximum temperature of chamber	Basal heat production per day			Respiratory quotient
					Total	Per kilogram of body weight	Per square meter of body surface	
Hens:		*Grams*	*Sq. cm.*		*Calories*	*Calories*	*Calories*	
E605	Nov. 9, 1925	1,925	1,551	63–70	126	65.5	812	0.70
	Nov. 17, 1925	1,880	1,533	62–80	132	70.2	861	.73
	Nov. 20, 1925	1,820	1,509	72–82	119	65.4	789	.71
	Dec. 4, 1925	1,715	1,464	72–82	100	58.3	683	.70
	Dec. 11, 1925	1,675	1,447	74–82	101	60.3	698	.70
	Dec. 14, 1925	1,630	1,428	72–78	88	54.0	616	.75
542	Sept. 28, 1925	1,790	1,496	70–82	109	60.9	729	.72
	Oct. 7, 1925	1,770	1,488	65–73	106	59.9	712	.71
	Oct. 10, 1925	1,740	1,475	63–71	102	58.6	692	.72
	Oct. 15, 1925	1,705	1,460	64–70	91	53.4	623	.71
	Oct. 18, 1925	1,645	1,434	54–62	90	54.7	628	.70
	Nov. 2, 1925	1,880	1,533	70–78	89	47.3	581	.73
	Nov. 5, 1925	1,880	1,533	71–76	91	48.4	594	.73
158 [a]	Jan. 29, 1923	1,657	1,379	67–75	94	56.7	682	.70
132 [a]	Feb. 1, 1923	1,630	1,359	74–78	81	49.8	595	.80
	Feb. 5, 1923	1,520	1,302	66–72	90	59.2	691	.70
	Feb. 8, 1923	1,525	1,304	72–80	82	53.8	629	.87
200	June 17, 1925	2,287	1,666	80–93	115	50.3	690	.71
	June 29, 1925	2,300	1,670	66–84	110	47.8	659	.71
	July 2, 1925	2,265	1,657	82–99	109	48.1	658	.71
	Sept. 7, 1925	1,990	1,554	80–89	103	51.8	663	.72
	Sept. 17, 1925	2,010	1,561	73–84	103	51.2	660	.70
	Sept. 20, 1925	1,990	1,554	69–79	92	46.2	592	.73
	Sept. 27, 1925	1,955	1,540	73–77	89	45.5	578	.72
	Oct. 11, 1925	1,895	1,516	62–66	102	53.8	673	.70
	Oct. 14, 1925	1,825	1,488	64–66	99	54.3	665	.65
A552	July 6, 1925	2,055	1,603	82–94	119	57.9	742	.70
	July 9, 9125	2,027	1,592	84–86	114	56.2	716	.72
2429 [a]	July 20, 1925	1,860	1,490	78–82	113	60.8	758	.70
	July 23, 1925	1,809	1,462	69–81	112	61.9	766	.71
546	Oct. 19, 1925	2,050	1,577	60–70	139	67.8	881	.71
	Oct. 25, 1925	2,020	1,565	68–74	120	59.4	767	.71
	Oct. 28, 1925	1,965	1,544	72–76	113	57.5	732	.72
	Nov. 3, 1925	1,900	1,518	67–75	110	57.9	725	.70
	Nov. 6, 1925	1,860	1,502	72–78	106	57.0	706	.70
35	Nov. 16, 1925	2,140	1,645	65–74	103	48.1	626	.71
	Nov. 23, 1925	2,115	1,636	58–72	102	48.2	623	.70
	Nov. 26, 1925	2,090	1,626	75–82	102	48.8	627	.72
	Dec. 4, 1925	2,100	1,632	74–84	85	40.5	520	.70
2420	Dec. 4, 1924	2,578	2,116	78–85	113	43.8	534	.72
2058	Dec. 11, 1924	2,195	1,759	80–92	122	55.6	694	.72
0	May 20, 1925	1,983	1,885	72–78	116	58.5	615	.69
2353	June 3, 1925	2,047	1,915	85–90	149	[b](72.8)	[b](778)	.70
2001	June 10, 1925	3,505	2,632	75–80	151	43.1	574	.69
Cocks:								
443 [a]	Aug. 25, 1924	2,750	1,933	78–88	148	53.8	766	.72
	Aug. 28, 1924	2,700	1,910	80–86	138	51.1	723	.69
458 [a]	Sept. 8, 1924	2,900	2,003	70–74	166	57.2	829	.70
	Sept. 11, 1924	2,800	1,957	72–82	154	55.0	787	.71
2284 [a]	Sept. 15, 1924	3,600	2,314	68–80	154	42.8	666	.72
	Sept. 18, 1924	3,450	2,249	68–76	166	48.1	738	.71
225 [a]	Sept. 22, 1924	3,235	2,155	68–82	191	59.0	886	.73
	Sept. 25, 1924	3,040	2,067	74–84	162	53.3	784	.72
161 [a]	Sept. 22, 1924	3,120	2,103	70–94	182	58.3	865	.71
	Sept. 25, 1924	2,945	2,024	73–93	173	58.7	855	.71
180 [a]	Aug. 18, 1924	2,330	1,731	73–84	127	54.5	734	.68
	Aug. 21, 1924	2,275	1,704	80–90	114	50.1	669	.72
552 [a]	Aug. 25, 1924	2,475	1,802	78–91	138	55.8	766	.73
	Aug. 28, 1924	2,410	1,771	72–88	131	54.4	739	.71

[a] The surface area of these birds was estimated by the Meeh formula (*9*), taking $k=9.85$. The surface area of all other birds was estimated from the weight and the rump-to-shoulder distance, according to the formula

$$S=5.86\ W^{0.5}\ L^{0.6}$$

of Mitchell, Card, and Hamilton (*11, p. 84*). Although this formula was obtained from White Plymouth Rock birds, it was shown to apply also to Rhode Island Red hens.

[b] Obtained during a period of egg production.

On the first experimental day these three Rhode Island Red hens were given a feeding of 75 gm. of ground corn before being placed in the respiration chamber; the heat production was then determined on five successive days in observational periods of 7 to 8 hours, the results being expressed on the 24-hour basis. In the case of hen 2353 the basal level was apparently established in 24 hours, but the fact that she was producing eggs possibly kept her heat production at a high level on the third and fourth days. The basal level was not reached with hen 200 and, less certainly with hen 2001, until they had fasted 48 hours. Many other experiments to be cited later support the conclusion that a hen can not be considered to be in the postabsorptive state until 48 hours have elapsed since the last feeding.

Before considering the experiments dealing with the heat increment due to feeding it is interesting to consider the results obtained on the basal heat production of Rhode Island Red chickens. Such data, collected from the feeding experiments, are reported in Table 5. The values obtained refer to the heat production after a 48-hour fast, observed in periods of 19 to 23 hours and expressed on the 24-hour basis. The results have also been expressed in calories per day per kilogram of body weight and per square meter of body surface (skin area).

Where more than one basal metabolism determination has been made upon a chicken the results have been arranged in the order in which they were obtained. It will be noted that in general a decrease in basal metabolism occurs during the course of a series of observations, particularly well shown with hens 542, 546, and E605. This decrease is probably comparable to that observed by Lusk and Du Bois (*8*) in dogs during a series of laboratory experiments and seems to be due to confinement and the resulting lowering of vitality. While there may be some suspicion that some of the results obtained are larger than the actual basal values because of exposure for short periods at night to environmental temperatures below the critical, the large majority of the heat productions do not appear to be complicated by this factor.

A number of other basal metabolism data on Rhode Island Red chickens are available from studies in which, for one reason or another, the heating effect of corn was not satisfactorily determined. The majority of these results were obtained at a time when the method of studying the effect of feeds on metabolism was being developed, and represent either initial determinations or, as in most cases, determinations after a three-day experimental period. They have been assembled in Table 6.

The homogeneity of the results given in Tables 5 and 6 is somewhat disturbed by the fact that the basal metabolism of the chickens in general decreased with the time spent on experiment. Hence, the average for a hen kept on experiment for a long period of time would not be comparable to that of a hen on experiment for a short period of time. A more significant average would, therefore, be obtained from the results secured in the first metabolism experiments on the birds, representing, as nearly as possible, their basal metabolism at the time of removal from the poultry farm.

TABLE 6.—*Basal heat production of a group of Rhode Island Red chickens*

Bird No.	Date	Body weight	Body surface [a]	Minimum and maximum temperature of chamber	Basal heat production per day			Respiratory quotient
					Total	Per kilogram of body weight	Per square meter of body surface	
Hens:		*Grams*	*Sq. cm.*		*Calories*	*Calories*	*Calories*	
230	Oct. 23, 1924	1,937	1,531	74–92	114	58.9	745	0.72
2217	Oct. 9, 1924	2,720	1,919	78–88	128	47.1	667	.72
2089	Oct. 16, 1924	2,490	1,810	74–84	127	51.0	702	.71
137	do	2,330	1,731	70–80	117	50.2	676	.71
91	Oct. 23, 1924	2,025	1,577	70–89	116	57.3	736	.76
2429	Oct. 30, 1924	1,940	1,532	77–87	117	60.3	764	.74
2076	do	2,132	1,632	76–86	124	58.2	760	.73
2053	Nov. 6, 1924	2,300	1,716	76–86	129	56.1	752	.72
2013	Nov. 13, 1924	2,295	1,714	76–84	130	56.6	759	.72
2160	do	2,242	1,687	76–86	113	50.4	670	.72
2210	Nov. 26, 1924	1,845	1,482	68–85	84	45.5	567	.87
2353	do	1,705	1,406	70–93	106	62.2	754	.72
200	Dec. 4, 1924	2,445	1,788	72–90	115	47.0	643	.74
2009	Dec. 11, 1924	2,405	1,768	74–90	119	49.5	673	.71
127	Jan. 26, 1923	2,138	1,635	60–78	101	47.2	618	.73
73	Feb. 12, 1923	2,575	1,851	66–82	160	62.1	864	.72
Cocks:								
174	Sept. 1, 1924	2,475	1,802	72–84	150	60.6	832	.67
	Sept. 4, 1924	2,400	1,766	74–82	124	51.7	702	.72
816	Sept. 1, 1924	2,812	1,962	76–88	187	66.5	953	.72
	Sept. 4, 1924	2,700	1,910	73–78	164	60.7	859	.71
328	Sept. 11, 1924	2,820	1,966	71–77	151	53.5	768	.72
316	Sept. 15, 1924	3,290	2,179	66–90	228	69.3	1,046	.70
	Sept. 18, 1924	3,085	2,087	68–80	196	63.5	939	.69
524	Oct. 2, 1924	3,125	2,105	68–90	178	57.0	846	.72
541	do	2,740	1,929	70–86	141	51.5	731	.73
721	July 21, 1924	2,475	1,803	76–88	146	59.0	810	.73
	July 24, 1924	2,400	1,766	81–82	112	46.7	634	.73
398	July 31, 1924	2,650	1,886	80–88	131	49.4	695	.68
474	Aug. 7, 1924	3,000	2,049	76–88	148	49.3	722	.73
524	Aug. 11, 1924	3,030	2,063	77–84	160	52.8	776	.68
	Aug. 14, 1924	2,970	2,035	72–80	155	52.2	762	.70
427	Aug. 11, 1924	3,010	2,054	72–78	158	52.5	769	.69
	Aug. 14, 1924	2,960	2,031	72–82	147	49.7	724	.71
446	Aug. 18, 1924	3,185	2,132	72–82	180	56.5	844	.68
	Aug. 21, 1924	2,900	2,003	82–90	186	64.2	929	.71

[a] The surface area of these birds was estimated by the Meeh formula (*9*), taking $k=9.85$.

Considering only the first successful determinations made upon the birds, preceded at most by three days' confinement in the respiration apparatus, the average basal heat production for the 28 hens was 54.9 calories per day per kilogram of body weight, or 703 calories per day per square meter of body surface. For the 19 cocks the averages were 55.7 calories and 806 calories, respectively. These results indicate a distinct sex difference in basal metabolism, in agreement with work upon other animals. The standard deviations of the individual determinations were 6.64 calories per kilogram body weight, and 81.5 calories per square meter body surface for the hens and 5.76 calories and 88.2 calories for the cocks. The coefficients of variation were 12.1 per cent on the body weight and 11.6 per cent on the body surface in the case of the hens and 10.3 per cent on the body weight and 10.9 per cent on the body surface in the case of the cocks. These coefficients of variation may be compared with those computed by Harris and Benedict (*6*) for human subjects. For men the coefficients found were 9.36 for the basal metabolism referred to weight and 8.05 for the basal metabolism referred to surface. For women the coefficients were 14.14 and 9.17, respectively.

The average respiratory quotient in the experiments reported in Table 5 was 0.713, and in the experiments reported in Table 6, 0.719. Evidently the metabolism of the chicken in the period from the forty-eighth to the seventieth hours after feeding is almost entirely at the expense of fat. While a small minority of the individual quotients were below the respiratory quotient for fat, such deficits are quite probably due to slight errors in the oxygen determinations and are so considered in the calculation of the heat production. The writers did not obtain the consistently low values for the quotient during fast reported by Gerhartz (*2*) in experiments on hens. The results of the broader but contradictory experience of the writers, as well as the inherent improbability of the general occurrence of conditions that would force the respiratory quotient to 0.65, 0.60, or lower values in short periods of fast, may well indicate that these low values are due to technical errors.

The basal heat production of hens just considered refers to the nonlaying hen. The only comparable values that have been found in the literature are those of Gerhartz (*2*) upon two hens. The basal heat production in three experiments for one of these hens, in a period of sexual inactivity, 13 to 23 hours after feeding, was found to be 584, 681, and 676 calories daily per square meter of body surface as computed by the Meeh formula (*9*) where $k = 9.49$. In the brooding period this hen gave values of 785 and 779 calories and another hen values of 917 and 795 calories. All of these values are quite within the range of variation found in the writers' experiments. Hári (*4*) has reported average basal heat production values of 698 and 1.036 calories per day per square meter of body surface for two geese; and Hári and Kriwuscha (*5*) values of 735 and 901 calories for two ducks.

According to Gerhartz, the hen producing eggs has a much larger basal metabolism than the nonlaying hen, a difference (increase) of 44 per cent being noted in one hen. Although the writers obtained their data largely from nonlaying hens, occasionally an egg was laid during an experimental observation. In Table 4 the egg production of hen 2353 apparently increased her basal heat production, since at the end of 48 hours of fast it amounted to 72.8 calories per day per kilogram, or 778 calories per day per square meter of body surface. (See Table 5.) The apparently large heating effect of 75 gm. of corn with hen 200 (Table 4) is also plausibly explained by the production of an egg on the second day of the experiment. On the other hand, in experiment 21 on hen A552 (see Table 9) the production of an egg on the seventh day of the experiment did not appear to disturb the level of heat production, nor was the estimated heating effect of the corn excessive, even though the basal value, 716 calories per day per square meter of surface, was near the average for all of the hens. The subject evidently requires further investigation, particularly since there is no obvious reason why egg production in the hen, any more than milk production in the cow, should increase the basal metabolism and the maintenance requirement.

THE HEATING EFFECT OF CORN

The corn used in the respiration experiments to determine the heat increment due to feeding was analyzed from time to time, with the results shown in Table 7.

TABLE 7.—*Chemical composition of corn used in respiration experiments*

Sample No.	Percentage of—						Gross energy (calories per gram)
	Dry matter	Crude protein	Nitrogen-free extract	Crude fiber	Fat	Ash	
1	91.2	10.0	73.9	2.68	3.27	1.38	4.10
2	92.4	8.3	76.1	2.99	3.70	1.35	4.10
3	92.1	9.7	74.1	2.86	3.99	1.39	4.13
4	87.9	9.0	73.2	2.18	2.19	1.33	3.78
5	89.9	8.8	73.5	2.69	3.74	1.19	3.99
Average	90.7	9.16	74.2	2.68	3.38	1.33	4.02

In view of the small variation among these analyses, particularly with reference to dry matter, it was not considered necessary to relate definite samples with definite experiments. The respiration experiments to be described may therefore be considered to relate to corn containing 91 per cent of dry matter and 4.02 calories of gross energy per gram.

In the first series of experiments on the heating effect of corn the plan was to determine the basal metabolism of the chicken after a 48-hour fast, to give 50 or 75 gm. of ground corn in one feeding at the beginning of the second experimental day, and to determine the 24-hour heat production on the day of feeding and on the two following days. The last day's result, obtained after 48 hours of fast, was considered to represent the basal metabolism. It was, in general, lower than the result of the first basal period. The increase in heat production due to the feeding of corn was computed by assuming that the decrease in basal metabolism, whenever such occurred, was linear. Where the variation in the weight of the bird was considerable this factor also was considered in estimating the basal metabolism on the day of feeding and on the following day. The total heating effect per 100 gm. of corn was then computed as the final result in each experiment. Before each day's experiment the chicken was given 50 c. c. of water by pipette. The results of 20 experiments of this type upon 15 Rhode Island Red chickens are summarized in Table 8.

TABLE 8.—*Heating effect of corn on mature chickens (one feeding)*

Experiment No.	Bird No.	Day of test	Weight of bird	Treatment	Minimum and maximum temperature of chamber	CO_2 given off per 24 hours	O_2 per 24 hours	Respiratory quotient	Heat production per 24 hours	Estimated basal heat production	Effect of corn	
											Observed	Calculated to 100 grams of corn
Hens:			*Gms.*			*Gms.*	*Gms.*		*Calories*	*Calories*	*Calories*	*Calories*
1	546	1	2,020	Fasted 48 hours	68–74	35.9	36.5	0.71	120			
		2	2,030	Fed 75 gm. corn	62–70	60.7	42.6	1.03	151	118	33	
		3	2,020	Fasted	70–74	38.7	37.6	.75	125	116	9	56
		4	1,965	do	72–76	34.0	34.4	.72	113			
2	546	1	1,900	Fasted 48 hours	67–75	32.3	33.4	.70	110			
		2	1,895	Fed 50 gm. corn	68–78	49.8	37.3	.97	131	109	22	
		3	1,895	Fasted	70–74	34.8	34.1	.74	113	108	5	54
		4	1,860	do	72–78	31.1	32.2	.70	106			
3	E605	1	1,880	Fasted 48 hours	62–80	39.9	39.9	.73	132			
		2	1,880	Fed 75 gm. corn	70–78	59.6	43.6	.99	154	128	26	
		3	1,875	Fasted	72–78	39.6	37.7	.76	126	123	3	40
		4	1,820	do	72–82	35.4	36.4	.71	119			
4	E605	1	1,675	Fasted 48 hours	74–82	29.5	30.8	.70	101			
		2	1,675	Fed 50 gm. corn	72–82	42.3	31.7	.97	111	97	14	
		3	1,670	Fasted	73–85	29.8	27.7	.78	93	92	1	30
		4	1,630	do	72–78	27.3	26.6	.75	88			
5	542	1	1,705	Fasted 48 hours	64–70	27.1	27.8	.71	91			
		2	1,710	Fed 50 gm. corn	62–65	41.0	30.7	.97	108	91	17	
		3	1,690	Fasted	58–66	29.8	28.0	.77	94	90	4	42
		4	1,645	do	54–62	26.2	27.3	.70	90			
6	542	1	1,880	Fasted 48 hours	70–78	26.9	26.9	.73	89			
		2	1,930	Fed 75 gm. corn	69–79	47.7	34.9	.99	123	90	33	
		3	1,945	Fasted	70–80	38.2	31.6	.88	108	90	18	68
		4	1,880	do	71–76	27.9	27.6	.73	91			
7	158	1	1,657	Fasted 48 hours	67–75	27.2	28.6	.70	94			
		2	1,686	Fed 75 gm. corn	66–80	42.4	31.5	.98	111	90	21	
		3	1,700	Fasted	74–82	40.1	28.4	1.03	100	86	14	47
		4	1,630	do	74–78	26.6	24.2	.80	81			
8	132	1	1,521	Fasted 50 hours	66–72	26.2	27.4	.70	90			
		2	1,553	Fed 75 gm. corn	72–76	41.2	31.0	.96	109	87	22	
		3	1,572	Fasted		38.5	29.4	.95	103	84	19	55
		4	1,525	do	72–80	28.9	24.1	.87	82			
9	200	1	2,300	Fasted 46 hours	66–84	31.5	32.1	.71	110			
		2	2,317	Fed 75 gm. corn	74–86	50.2	38.3	.95	134	110	24	
		3	2,330	Fasted	78–88	43.0	36.3	.86	124	110	14	51
		4	2,265	do	82–99	32.5	33.2	.71	109			
10	A552	1	2,055	Fasted 46 hours	82–94	34.9	36.3	.70	119			
		2	2,063	Fed 75 gm. corn	82–92	50.4	37.8	.97	132	117	15	
		3	2,065	Fasted	82–88	43.8	38.2	.83	130	116	14	40
		4	2,027	do	84–86	34.3	34.5	.72	114			
11	2429	1	1,860	Fasted 48 hours	78–82	33.1	34.4	.70	113			
		2	1,857	Fed 75 gm. corn	74–88	48.1	37.1	.94	129	112	17	
		3	1,864	Fasted	68–83	43.1	36.7	.86	125	112	13	40
		4	1,809	do	69–81	33.3	34.0	.71	112			
12	35	1	2,115	Fasted 48 hours	58–72	30.1	31.1	.70	102			
		2	2,140	Fed 75 gm. corn	76–80	52.1	36.4	1.04	129	102	27	
		3	2,135	Fasted	74–79	36.6	31.5	.84	107	102	5	43
		4	2,090	do	75–82	30.6	30.9	.72	102			
13	200	1	2,010	Fasted 48 hours	73–84	30.4	31.5	.70	103			
		2	2,035	Fed 75 gm. corn	80–87	51.4	36.2	1.03	129	99	30	
		3	2,040	Fasted	80–88	37.9	32.0	.86	109	95	14	59
		4	1,990	do	69–79	28.1	28.0	.73	92			
Cocks:												
14	443	1	2,750	Fasted 48 hours	78–88	44.5	45.1	.72	148			
		2	2,720	Fed 75 gm. corn	80–88	62.0	50.2	.90	173	145	28	
		3	2,760	Fasted	80–90	48.1	47.5	.74	157	142	15	57
		4	2,700	do	80–86	40.0	42.0	.69	138			
15	552	1	2,475	Fasted 48 hours	78–91	42.2	41.7	.73	138			
		2	2,445	Fed 75 gm. corn	82–90	64.2	52.3	.89	180	136	44	
		3	2,475	Fasted	77–93	42.4	42.2	.73	139	134	5	65
		4	2,410	do	72–88	39.0	39.8	.71	131			
16	458	1	2,900	Fasted 48 hours	70–74	48.8	50.7	.70	166			
		2	2,900	Fed 75 gm. corn	72–80	69.9	55.0	.92	191	162	29	
		3	2,845	Fasted	70–82	49.4	49.4	.72	163	158	5	45
		4	2,800	do	72–82	45.8	46.9	.71	154			
17	2,284	1	3,600	Fasted 48 hours	68–80	46.0	46.8	.72	154			
		2	3,515	Fed 75 gm. corn	72–84	70.3	54.4	.94	189	158	31	
		3	3,520	Fasted	66–80	53.8	51.8	.75	172	162	10	55
		4	3,450	do	68–76	49.5	50.5	.71	166			

TABLE 8.—*Heating effect of corn on mature chickens (one feeding)*—Continued

Experiment No.	Bird No.	Day of test	Weight of bird	Treatment	Minimum and maximum temperature of chamber	CO_2 given off per 24 hours	O_2 per 24 hours	Respiratory quotient	Heat production per 24 hours	Estimated basal heat production	Effect of corn	
											Observed	Calculated to 100 grams of corn
			Gms.			*Gms.*	*Gms.*		*Calories*	*Calories*	*Calories*	*Calories*
18	225	1	3,235	Fasted 48 hours	68–82	57.9	57.9	0.73	191			
		2	3,150	Fed 75 gm. corn	70–80	74.8	61.4	.89	211	181	30	
		3	3,100	Fasted	71–81	62.2	55.0	.82	186	171	15	60
		4	3,040	do	74–84	48.6	49.2	.72	162			
19	161	1	3,120	Fasted 48 hours	70–94	54.4	55.4	.71	182			
		2	3,080	Fed 75 gm. corn	66–78	76.1	65.6	.84	223	179	44	
		3	3,000	Fasted	70–76	62.0	57.4	.78	192	176	16	(93)
		4	2,945	do	73–93	51.8	52.7	.71	173			
20	180	1	2,330	Fasted 48 hours	73–84	39.2	38.6	.68	127			
		2	2,275	Fed 75 gm. corn	76–82	57.1	43.2	.96	151	123	28	
		3	2,275	Fasted	78–86	41.6	39.4	.77	131	119	12	53
		4	2,275	do	80–90	34.3	34.5	.72	114			

Another method of attacking the same problem is to compare the level of heat production established by the continuous feeding of a constant amount of corn with the basal heat production. In this type of experiment the basal metabolism was determined after 48 hours of fast, after which the chickens were given 50 or 75 gm. of ground corn and an adequate and constant amount of water early each morning. The heat production was determined daily until, for several consecutive days, a level appeared to have been established, following which food was withdrawn and a basal determination again made after a fast of 48 hours. The heating effect of the corn fed was then taken as the difference between the average heat production on feed and the average estimated basal heat production. Eleven experiments of this type upon five different Rhode Island Red hens are summarized in Table 9.

TABLE 9.—*Heating effect of corn on hens (continuous feeding)*

Experiment No.	Bird No.	Day of test	Weight of bird	Treatment	Minimum and maximum temperature of chamber	CO_2 given off per 24 hours	O_2 consumption per 24 hours	Respiratory quotient	Heat production per 24 hours	Average heat production on feed	Estimated basal heat production	Effect of corn	
												Observed	Calculated to 100 grams
			Grams			*Gms.*	*Gms.*		*Calories*	*Calories*	*Calories*	*Calories*	*Calories*
21	A552	1	2,027	Basal	84–86	34.3	34.5	0.72	114				
		5	2,210	Fed 75 gm. corn	77–91	57.2	42.7	.97	150				
		6	2,170	do	76–84	68.0	47.2	1.05	168				
		*7	2,157	do	84–88	58.4	44.6	.95	156				
		8	2,135	do	81–97	68.3	45.5	1.09	163	159	122	37	50
22	546	1	2,050	Basal	60–70	41.4	42.3	.71	139				
		3	2,095	Fed 75 gm. corn	64–86	71.9	48.4	1.08	173				
		4	2,100	do	68–87	69.3	47.0	1.07	168				
		5	2,105	do	69–85	68.6	46.5	1.07	166	169	133	36	48
		7	2,020	Basal	68–74	35.9	36.5	.71	120				

* Hen laid an egg (45 gm.) in respiration chamber.

TABLE 9.—*Heating effect of corn on hens (continuous feeding)*—Continued

Experiment No.	Bird No.	Day of test	Weight of bird	Treatment	Minimum and maximum temperature of chamber	CO_2 given off per 24 hours	O_2 consumption per 24 hours	Respiratory quotient	Heat production per 24 hours	Average heat production on feed	Estimated basal heat production	Effect of corn	
												Observed	Calculated to 100 grams
			Grams			*Gms.*	*Gms.*		*Calories*	*Calories*	*Calories*	*Calories*	*Calories*
23	546	1	1,965	Basal	72–76	34.0	34.4	0.72	113				
		3	1,950	Fed 50 gm. corn	68–76	53.7	40.4	.96	142				
		4	1,950	do	68–72	55.2	41.0	.98	144				
		5	1,955	do	70–76	55.9	40.6	1.00	143	143	112	31	62
		7	1,900	Basal	67–75	32.3	33.5	.70	110				
24	E605	1	1,925	do	63–70	37.2	38.3	.70	126				
		3	1,955	Fed 75 gm. corn	68–82	62.2	43.9	1.03	156				
		4	1,965	do	76–79	63.3	44.0	1.04	157				
		5	1,975	do	68–80	66.7	44.6	1.09	160				
		6	1,980	do	74–78	64.2	44.0	1.06	157				
		7	1,975	do	62–76	64.2	44.2	1.06	159	158	129	29	39
		9	1,880	Basal	62–80	39.9	39.9	.73	132				
25	E605	1	1,820	do	72–82	35.4	36.4	.71	119				
		3	1,840	Fed 50 gm. corn	54–72	56.0	40.6	1.00	143				
		4	1,830	do	54–77	51.7	40.0	.94	139				
		5	1,835	do	74–98	51.3	37.9	.98	134	138	119	19	38
		6	1,855	do	76–84	50.6	38.8	.95	135				
26	E605	1	1,715	Basal	72–82	29.3	30.5	.70	100				
		3	1,740	Fed 50 gm. corn	70–82	46.8	34.2	.99	121				
		4	1,755	do	70–84	48.7	34.6	1.02	123				
		5	1,750	do	76–84	47.5	34.6	1.00	122				
		6	1,745	do	74–94	49.6	35.6	1.01	126	123	101	22	44
		8	1,675	Basal	74–82	29.5	30.8	.70	101				
27	542	1	1,790	do	70–82	32.7	33.1	.72	109				
		6	1,835	Fed 75 gm. corn	68–75	50.5	40.1	1.00	142				
		7	1,830	do	65–75	60.9	39.3	1.13	141				
		8	1,830	do	68–76	59.9	40.1	1.09	143	142	108	34	45
		10	1,770	Basal	65–73	31.4	32.3	.71	106				
28	542	1	1,740	do	63–71	30.6	30.9	.72	102				
		2	1,730	Fed 50 gm. corn	63–73	44.3	33.6	.96	118				
		3	1,735	do	65–71	47.6	32.6	1.06	116				
		4	1,735	do	68–70	46.9	33.0	1.03	117	117	96	21	42
		6	1,705	Basal	64–70	27.1	27.8	.71	91				
29	35	1	2,140	do	65–74	30.5	31.4	.71	103				
		3	2,180	Fed 75 gm. corn	70–77	65.0	44.0	1.07	157				
		4	2,165	do	70–78	66.8	46.2	1.05	165				
		5	2,165	do	76–78	66.6	44.4	1.09	159				
		6	2,170	do	70–76	65.7	44.8	1.06	159	160	105	55	73
		8	2,115	Basal	58–72	30.1	31.1	.70	102				
30	200	1	1,990	do	80–89	31.0	31.4	.72	103				
		3	2,017	Fed 75 gm. corn	80–89	62.9	41.0	1.11	147				
		4	2,045	do	80–86	63.4	41.6	1.11	149				
		5	2,070	do	80–82	68.9	43.4	1.15	157				
		6	2,070	do	74–78	67.9	46.4	1.06	(165)				
		7	2,080	do	74–79	65.6	42.3	1.13	152				
		8	2,080	do	73–79	62.3	40.9	1.11	147				
		9	2,080	do	72–77	62.5	41.2	1.10	148	150	106	44	59
		11	2,010	Basal	73–84	30.4	31.5	.70	103				
31	200	1	1,990	do	69–79	28.1	28.0	.73	92				
		2	1,985	Fed 50 gm. corn	65–72	45.5	35.6	.93	123				
		3	2,000	do	65–70	54.2	38.2	1.03	(136)				
		4	2,000	do	65–75	49.7	35.3	1.02	125				
		5	2,000	do	62–76	49.9	35.6	1.02	126				
		6	2,000	do	62–73	49.0	34.7	1.02	123	124	91	33	66
		8	1,955	Basal	73–77	26.6	27.0	.72	89				

In considering these two sets of data it is well at the start to determine how homogeneous they are, particularly with respect to (1) the type of experiment and (2) the amount of food given. Disregarding the exceptionally high result of experiment 19 in the first series of experiments, there were 16 involving a feeding of 75 gm. of corn and only 3 involving a feeding of 50 gm. The average heating

effect of corn per 100 gm. was in the first case 52 calories and in the second case 42 calories, the average for the entire series being 50 calories. Among the second series of experiments there were 6 involving a feeding of 75 gm. portions of corn and 5 involving a feeding of 50 gm. portions. The average heating effects were, respectively, 52 calories and 50 calories per 100 gm. of corn; and for the entire group 51 calories. From these averages it may be concluded, in the first place, that the two types of experiments gave essentially the same results. In the early experiments the heating effect extended over two days; animals fed 75 gm. of corn excreted on an average 70 per cent of the extra heat on the day of feeding and 30 per cent on the day following, while animals fed 50 gm. portions excreted on an average 85 per cent the first day and only 15 per cent on the day following. In the second series of experiments, therefore, the level of heat production established by continuous feeding must have exceeded that established by one feeding by exactly the "carry-over" effect of the day following one feeding.

In the second place, the above averages indicate that the smaller portion of corn *may* have had a slightly smaller heating effect per 100 gm. than the larger, but it is evident from an inspection of the tables that the data are too few and too variable to establish the significance of the average differences obtained.

Hence, the data may be considered and treated as a homogeneous series as far as these possible vitiating factors are concerned. It is true that where several tests were made upon the same bird there is some indication of rather distinct individual differences, as the summary in Table 10 shows.

TABLE 10.—*Difference among individual birds in the heating effect of corn per 100 gm.*

Bird No.	Experiment No.	Heat increment per 100 gm. of corn	Bird No.	Experiment No.	Heat increment per 100 gm. of corn
546	1	56	542	5	42
	2	54		6	68
	22	48		27	45
	23	62		28	42
E605	3	40	A552	10	40
	4	30		21	50
	24	39	200	9	51
	25	38		13	59
	26	44		30	59
				31	66
			35	12	43
				29	73

The results obtained with hen 200 are quite sharply distinguished from those obtained with hen E605. Hen 546, except for one of the four experiments, gave higher results than the average, and hen 542, except for one result, gave lower figures than the average. These differences are undoubtedly a source of heterogeneity. However, in view of the number of results obtained and the many other uncontrolled factors that were evidently operating in determining the estimated heating effect of corn, the effect of these differences is probably not serious.

The results of seven experiments upon cocks are summarized in Table 8. Omitting the anomalous result of experiment 19, the average heating effect of corn was 56 calories per 100 gm. For the 13 results on hens the average was 48 calories. The slight difference may be of significance in indicating a smaller heating effect for hens, but the data are too few and variable to permit a definite conclusion on this interesting point.

It is necessary to inquire whether the low minimum temperatures of the animal chamber prevailing at some period during the night in several of the experiments might not have operated in vitiating the estimated heat effect of corn on the metabolism of the chickens. The effect of temperatures below the critical would be to raise the heat production above the level that would otherwise prevail. For any low temperature the effect, if existent, would be more marked in the basal periods than in the feeding periods, since the critical temperature in the basal periods would be higher. Hence, where low temperatures exert such a vitiating effect the result would always be to depress the estimated heat increment below the actual. It is extremely doubtful whether on any of the feeding days the temperature in the chamber ever was less than the critical temperature of the fed bird.

It has been shown elsewhere by the writers (*10*) that the average critical temperature of the fasting Rhode Island Red hen is 62° F., though individual hens may show considerable variability in this respect. There were six experiments in which at least one of the two basal periods possessed a minimum temperature of 62° or below. The average estimated heat increment for these six experiments was 47, as compared with the average of 51 for the entire investigation. Among the six results is included the highest value, 73, obtained in this study, disregarding the anomalous result of experiment 19. The two lowest found, 30 and 38 (experiments 4 and 25), were obtained under temperature conditions above suspicion.

In this connection a further study of Table 10 is of interest. The lowest result for hen 546 is open to suspicion because of a minimum temperature of 60° F. in one of the basal periods, while for hen 542 the three low results may be considered suspicious for the same reason. However, for hen 35 no such explanation of the low value is plausible, while the relative constancy in results for hens E605 and 200 was obtained in spite of a wide variety of temperature conditions.

While it appears, therefore, that some of the low estimates of the heating effect of corn may have been occasioned in part by the prevalence during a fraction of the basal periods of temperatures below the critical, it may readily be shown that such temperature effects were not of serious moment. Eliminating the results of experiment 19 and of 6 experiments in which, for at least one of the basal periods, the minimum temperature was 62° F. or below, gives an average result for the remaining 24 experiments of 52 calories per 100 gm. of corn. If 8 experiments in which, for at least one of the basal periods, the minimum temperature was 65° or below are disregarded, the average result of the remaining 22 experiments is 52 calories per 100 gm. of corn. If the process of elimination is carried further to include all experiments in which the minimum temperature was 70° or below for at least one of the basal periods the average of the remaining 9 experiments becomes 48 calories.

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The average heating effect of corn for all 30 experiments (excluding No. 19) is 50.7 ± 1.2 calories per 100 gm. of corn. The standard deviation of the individual determinations is 10.04 calories, and the coefficient of variation 19.8 per cent.

THE NET-ENERGY VALUE OF CORN FOR MATURE CHICKENS

Summing up the results of the investigation, it has been found that 83 per cent of the gross energy of ground corn is metabolizable by chickens and that the average heating effect of corn on cocks and nonlaying hens is 51 calories per 100 gm. The corn used in these investigations contained 402 calories per 100 gm. Its metabolizable energy would, therefore, be 334 calories and its net energy $334 - 51 = 283$ calories per 100 gm., or 1,285 calories per pound. Since the corn used in this investigation contained an average of 91 per cent of dry matter, the net-energy value of corn is 3,110 calories per kilogram of dry matter, or 1,412 calories per pound of dry matter. The latter figure may be compared with the value Armsby gives (*1, p. 660, 722*) for cattle, 926 calories per pound of dry matter, and his estimate for swine, i. e., 1,355 calories. Evidently the nonlaying hen ranks high in her ability to convert the energy of corn to her own uses.

SUMMARY

In experiments upon 10 chickens it has been found that 83 per cent of the gross energy of ground corn is metabolizable.

The average basal heat production of 28 nonlaying hens was found to be 54.9 calories per day per kilogram body weight and 703 calories per day per square meter body surface. For 19 mature cocks the average values found were 55.7 and 806 calories, respectively. These values may be considerably reduced during protracted confinement in the laboratory. The basal metabolism of hens is apparently raised considerably during periods of egg production, though this question requires much further study.

In 30 experiments upon 15 mature Rhode Island Red chickens an average heating effect of 50.7 ± 1.2 calories per 100 gm. of corn (91 per cent dry matter) has been obtained. After a single feeding of 75 gm. of ground corn an average of 70 per cent of the extra heat is eliminated during the first 24 hours and 30 per cent during the second 24 hours. For a single feeding of 50 gm. of corn 85 per cent of the extra heat is excreted in 24 hours and 15 per cent in the second 24-hour period. The chicken, therefore, can not be considered to be in the postabsorptive condition until more than 24 hours have elapsed since the last feeding. After 48 hours this condition may be considered to have been established. Some evidence was obtained indicating that the heating effect of corn is slightly greater with cocks than with hens.

The net-energy value of corn for mature chickens has been found to be 3,110 calories per kilogram of dry matter, or 1,412 calories per pound of dry matter.

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THE EFFECT OF AGE, SEX, AND CASTRATION ON THE BASAL HEAT PRODUCTION OF CHICKENS[1]

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INTRODUCTION

The basal heat production of an animal is its minimum energy expenditure. This factor determines the minimum requirement of food energy, and appears to be one of the most constant of biological measurements. When dividing an animal's energy expenditures into independent factors, the basal heat production is the factor upon which all others are imposed, and it becomes, therefore, the logical starting point in experimental investigations of this nature. Its intimate relation to the maintenance requirement of energy, of which it is the most important part, gives it a practical as well as a scientific significance.

A study of the basal heat production of chickens should start with the incubating egg if the developing embryo could be reduced to the fasting condition. The metabolism of the embryo within the egg involves the basal expenditure of energy and also whatever calorigenic effect the absorbed nutrient material may exert upon the embryonic tissue. It is an interesting fact, however, that the carbon dioxide production of the developing chick increases in proportion to its weight (*9, 10, 11, 20, 21*)[2] and that the heat produced, except possibly during the first few days, is essentially at the expense of fat.

During its life within the egg, the chick is cold-blooded (*35*), except for the few hours preceding hatching, when the transformation from the cold-blooded to the warm-blooded state occurs. In other words, during incubation the heat production of the embryo varies directly with its environmental temperature, responding to a fall in temperature with a fall in heat production, and vice versa. The writers have been able to confirm the results of Pembrey, Gordon, and Warren on this point.[3] However, during its first day or so of life, the

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[2] Reference is made by number (italic) to "Literature cited," p. 958.

[3] Two eggs showed the following responses to temperature changes, one on the 19th and one on the 20th day of incubation:

Day of incubation	Temperature of receptacle	CO_2 produced per 30 minutes	Day of incubation	Temperature of receptacle	CO_2 produced per 30 minutes
	° *C.*	*Mgm.*		° *C.*	*Mgm.*
Nineteenth	40	14.0	Twentieth	40	12.4
	40	15.2		40	13.8
	40	15.1		25	7.5
	25	7.6		40	14.5
	40	16.2		40	14.5
	40	13.8		40	14.8
	25	7.8		25	7.9

chick quickly assumes the characteristics of warm-bloodedness. It becomes less a creature of circumstance. Throughout a considerable temperature range, its metabolism is unaffected; its body temperature is maintained constant at varying environmental temperatures by physiological regulation of the emission of heat from its body. When this regulatory mechanism is strained to the utmost by decreasing temperatures, it is still able to maintain its body temperature by increasing its heat production, the direct opposite of its response intra ovum.

In its changing response to environmental temperatures previous and subsequent to hatching, the embryo is reminiscent of the hibernating animal. During hibernation, the response of an animal is typically cold blooded. It lacks the capacity of maintaining its body temperature constant, and its metabolism, in the prevailing cold weather, decreases, often to a mere fraction of that which characterizes its active existence. When environmental conditions, particularly with reference to temperature, become suitable, the animal wakes from hibernation, there is an outburst of metabolic activity and a rapid increase in heat production, and as a result the body temperature quickly rises to normal (*3*, *17*, *18*, *31*, *32*, *33*, *34*, *36*).

A similar outburst of metabolic activity occurs in the hatching chick, although not of the same relative magnitude. Thus, the carbon dioxide production of an embryo on the nineteenth day of incubation was 29 mgm. per hour, and that of another embryo on the twentieth day was 28 mgm. per hour. On the other hand, a chick of the same breed 1 day old, when quiet and fasting, produced 68 mgm. of CO_2 per hour; another chick, 2 days old, produced 61 mgm. per hour, while a third chick, 3 days old, produced 61.4 mgm. Since the respiratory quotient of the incubating embryo is similar to that of the fasting chick, the rates of heat production of the two stand in the same ratio as the rates of production of carbon dioxide. The experimental observations of Pembrey, Gordon, and Warren (*35*) also indicate a marked increase in the metabolism of the chick at hatching.

The effect of age and sex upon the basal metabolism has not been studied in any great detail except with human subjects. In 1916, Du Bois (*15*) presented a curve showing the change in the basal metabolism per unit of body surface from birth to old age. During the first few weeks of life the metabolism of the infant was found to be very low (27 to 30 calories per square meter per hour); it then rose rapidly until at 1 year of age it was 50 per cent above the adult level. The maximum was reached in the "almost unexplored" period between 2 and 6 years of age (about 60 calories per square meter per hour), after which the metabolism fell rapidly to 20 years of age, and extremely gradually after that. The essential features of this curve have been confirmed by Benedict and Talbot (*6*, *8*), and especially by the more recent data of Talbot (*37*). Mitchell and Carman (*27*) observed a very constant rate of basal metabolism in rats from 90 to 190 days of age, though at an earlier age (30 to 40 days) distinctly higher rates were noted. Essentially the same effect of age upon basal metabolism was observed in growing pigs by Deighton (*14*) and Wood (*38*).

It is generally recognized that females possess a lower rate of basal metabolism than males (*4*, *7*, *19*), and with human subjects, accord-

ing to Benedict (*6*, *7*), the sex difference appears at an early age, represented by a body weight of 10 kgm. In the human species, the difference in basal metabolism between the sexes is ordinarily rated at 5 to 7 per cent.

EXPERIMENTAL METHODS

In the writers' studies of the effect of age and sex upon basal heat production, the Haldane gravimetric method of determining the respiratory exchange of small animals (*16*) was used. The apparatus employed has been described in an earlier publication from this laboratory (*28*). For different-sized chickens different-sized chambers were employed, the endeavor being to fit the chamber snugly to the bird so as to minimize the possibility of considerable muscular activity and yet to permit the chicken to assume a comfortable posture.

In the earlier work, performed in 1923, no method of recording the activity of the birds had been elaborated, but in the later experiments (*29*) the animal chamber was suspended from a spring and the vertical movement of the chamber was recorded on a revolving "work-adder." The records thus obtained, which appear in Tables 4, 5, and 6 as "work record," are purely of comparative significance.

The heat production was computed by the well-known method of Zuntz and Schumburg (*25*, *p. 56–63; 39*), upon the assumption that the total respiratory quotient is nonprotein, thus neglecting the protein metabolism. From the available data relating to the calorific value of oxygen used in the combustion of the different food nutrients, it may be computed that the maximum error in the estimate of the total heat production resulting from this simplification is 6.58 per cent, while the usual errors would be less than 2 per cent.

EXPERIMENTAL DATA

The experimental results will be considered, not in the order in which they were obtained, but with reference to the age of the chicks. For this reason, some of the results to be considered later will be less satisfactory than some of those considered first, because of a better control of experimental conditions as familiarity with the methods increased.

The experiments with birds just after hatching were performed upon White Plymouth Rocks. The chicks were confined in small Mason jars and were under observation for three to seven hours, during which the respiratory quotient was determined. The CO_2 production during two or more 30-minute periods of quietness was found, and upon these values the basal heat production was computed. The surface area of the birds was estimated on the basis of 10 determinations of the skin area of as many White Plymouth Rock chicks 1 day old. The average weight of these chicks was 36 gm. and their average skin area, 91 sq. cm. In estimating the area of the experimental chicks, it was assumed that it would vary in proportion to the two-thirds power of the body weight. The average data from these tests are given in Table 1.

TABLE 1.—*The basal heat production of young White Plymouth Rock chicks*

Chick No.	Age	Condition	Body weight	Body surface	Temperature of chamber	O_2 consumed daily	Respiratory quotient	Heat production per day	
								Total	Per square meter body surface
	Days		*Gm.*	*Sq. cm.*	*° C.*	*C. c.*		*Cals.*	*Cals.*
1	1	Fasted since hatching	35.6	91	36.0	1,008	0.82	4.87	535
2	2	do	30.7	78	37.5	871	.86	4.24	544
3	3	do	32.9	83	37.5	959	.78	4.57	551
4	5	Fasted 36 hours	31.2	79	37.5	1,270	.73	5.97	756
5	6	Fasted 60 hours	31.3	79	37.5	934	.86	4.56	577
6	6	Fasted 65 hours	30.8	79	37.5	1,061	.90	5.22	661
7	13	Fasted 44 hours	25.3	64	37.5	899	.78	4.28	669
8	15	do	31.7	80	37.5	1,323	.83	6.39	799
9	16	Fasted 25 hours	30.8	78	37.5	1,354	.80	6.49	832
10	17	Fasted 43 hours	30.3	77	37.5	1,343	.75	6.35	825

Omitting the exceptional result for chick 4, the heat production per square meter of body surface increased regularly from the day-old chick to the 16-day-old chick. In an earlier publication (*28*) an average heat production of 703 calories per day per square meter of surface was observed with 28 mature Rhode Island Red hens, and one of 806 calories with 19 mature cocks. Evidently the chick immediately after hatching has a basal heat production considerably below the level of the mature chicken. These results are quite similar to those reported for infants.

In the next group of experiments concerned with the relation between age and basal heat production, young single-comb White Leghorn chicks fasted for 48 hours, were used. An attempt was made to reduce the activity of these birds to a minimum by wrapping them in cheesecloth, except for the head; the legs (in the squatting or roosting position) were bound somewhat loosely to the body. Two 3-hour periods were run upon each chick, one in the morning and one in the afternoon. In practically all cases, the production of heat was considerably higher in the morning than in the afternoon, the difference being the more marked with the younger chicks. In most cases where the rectal temperatures were observed, a distinct reduction of body temperature was noted from the time the chick was put in the chamber in the early morning to the time it was removed in the late afternoon. The exceptions were related generally to excessively high chamber temperatures in the afternoon, which were reflected in rectal temperatures of 108° to 110° F. It was at first thought that this reduction in metabolism was a diurnal phenomenon, and the procedure was therefore reversed with six chicks, the first experimental period being in the afternoon and the second in the morning following. In four of these experiments the morning heat production exceeded the afternoon, but in two cases the reverse was true.

The differences observed between morning and afternoon results (averaging 21 per cent with the youngest group and 12 per cent with the oldest) seemed too large to be a diurnal phenomenon, if one may judge from results on human subjects (*5*), and, moreover, they seemed to be in the wrong direction—with men the higher level appears to be in the afternoon. A more probable explanation is that

the difference was due to muscular activity. Quite possibly in the morning the chick struggled to some extent against its bandage, but in the afternoon had become either exhausted or reconciled to the experimental conditions. One experiment in which the bird was unwrapped in the morning but wrapped in the afternoon confirms this conclusion. During the morning the CO_2 excretion during a period of quietness (37 minutes) was measured, from which, in conjunction with the respiratory quotient for the three-hour period, it was computed that the heat production was at the rate of 10.38 calories per day. The value for the entire afternoon period, during which the bird was wrapped in cheesecloth, was 15.57 calories per day. This value expressed per square meter of body surface (1,188 calories) is clearly out of line with the other afternoon values for chicks of approximately the age of this one (13 days). It was therefore decided to discard the morning values in all of these experiments and to consider the lower afternoon values as more nearly approximating the basal heat production.

TABLE 2.—*The basal heat production of young single comb White Leghorn chicks*

Chick No.	Sex	Age	Body weight	Body surface	Body temperature	Maximum and minimum temperatures of chamber	O_2 consumed daily	Respiratory quotient	Heat production per day	
									Total	Per square meter body surface
		Days	*Gm.*	*Sq. cm.*	*°F.*	*°F.*	*Liters*		*Cals.*	*Cals.*
4776	Male	7	40.0	102		85–95	1.46	0.73	6.86	675
4540	do	8	43.4	107		85–95	1.85	.70	8.66	807
4761	do	9	38.7	99		85–95	1.23	.76	5.85	591
4733	Female	12	49.7	117		85–95	1.96	.75	9.28	790
4749	Male	13	58.2	130		85–95	2.19	.74	10.38	796
4953	do	14	60.8	134		85–95	1.46	.81	7.00	521
4891	do	16	62.4	137		85–95	1.85	.73	8.70	637
4505	do	19	86.0	169		85–90	3.92	.75	14.57	861
4762	do	20	76.8	157		85–95	3.64	.70	17.06	1,087
4947	do	21	82.9	164		85–95	4.07	.71	19.11	1,162
4604	do	22	71.8	150		87–92	3.19	.71	14.86	907
4849	do	23	94	179		85–95	4.03	.71	18.89	1,052
4670	do	27	126	218		80–90	4.82	.73	22.68	1,040
4628	do	29	147	242		80–85	7.34	.72	34.46	1,423
4387	do	30	128	220		80–88	4.70	.73	22.15	1,004
5000	do	98	730	932	106.4	92–94	13.66	.80	65.7	704
4717	do	104	820	1,008	106.4	89–92	14.28	.72	67.2	667
4900	do	106	1,010	1,180	106.4	92–95	19.38	.71	91.0	771
	Average									714
4511	Female	97	770	915	106.2	88–92	15.79	.73	74.4	813
4945	do	99	740	885	108.0	90–93	14.22	.73	67.1	758
4969	do	103	610	760	107.2	87–92	14.22	.73	67.0	881
4521	do	105	730	875	107.2	89–93	14.28	.76	67.9	776
4933	do	107	800	940	105.8	87–91	15.74	.72	67.0	713
	Average									788
4885	Male	146	1,500	1,580	104.8	72–80	25.41	.71	119.3	755
4832	do	149	800	995	105.0	70–78	14.50	.75	68.8	691
4564	do	152	1,390	1,530	105.0	70–76	27.05	.70	126.7	828
4755	do	154	1,600	1,620	104.8	76–84	22.96	.73	108.3	669
4865	do	161	1,390	1,530		78–84	19.54	.71	91.6	599
4900	do	163	1,650	1,640	106.0	76–82	26.66	.74	126.2	770
	Average									718
4858	Female	147	1,320	1,310	105.2	72–80	21.50	.71	100.8	769
4739	do	148	1,235	1,263	104.8	74–78	18.93	.71	88.8	703
4964	do	153	1,170	1,225	105.5	70–78	19.71	.70	92.4	754
4992	do	160	1,230	1,260	104.2	72–78	16.18	.71	76.0	603
4553	do	166	1,380	1,340	104.7	72–78	25.54	.70	119.7	890
	Average									744

TABLE 3.—*The average skin area of single comb White Leghorn chickens of different sex; each area is the average for 10 birds*

Body weight of cockerels	Average skin area of cockerels			Body weight of pullets	Average skin area of pullets		
	Body	Combs and wattles[a]	Total		Body	Combs and wattles[a]	Total
Gm.	*Sq. cm.*	*Sq. cm.*	*Sq. cm.*	*Gm.*	*Sq. cm.*	*Sq. cm.*	*Sq. cm.*
[b]31.1	86	----------	86	----------	----------	----------	----------
218	294	----------	294	222	325	----------	325
477	568	----------	568	468	602	----------	602
678	840	36	876	669	815	----------	815
874	986	66	1,052	890	1,016	----------	1,016
1,317	1,367	116	1,483	1,367	1,298	34	1,332
1,719	1,536	124	1,660	1,716	1,350	72	1,422
2,136	1,689	160	1,849	----------	----------	----------	----------

[a] The areas given are twice the areas outlined.
[b] The sex of these birds was not determined.

A summary of the data in this series of experiments is given in Table 2. In estimating the surface area of these chicks, advantage was taken of a considerable number of determinations of the skin area of single comb White Leghorn chickens made in the course of another investigation. In this investigation, the skin area of 10 chicks 1 day old was determined, and thereafter the skin areas of groups of 10 cockerels and 10 pullets at the approximate weights of 0.5, 1, 1.5, 2, 3, 4, and 5 pounds, except that no 5-pound pullets were measured. The areas refer to the body only, the skin of the shanks and feet not being determined. For the older birds, with combs and wattles of appreciable size, the area of these appendages was determined and multiplied by 2 to get their approximate surface area. The average skin areas of the different groups of chickens are given in Table 3. These areas were plotted upon coordinate paper against the body weights, and a separate curve was made for cockerels and pullets; the areas of the experimental birds was then estimated graphically from their body weights. It will be noted that with this group of birds, as with the group of young White Plymouth Rocks, the basal heat production per square meter of body surface starts at a low value and increases progressively. For these White Leghorns, the maximum basal metabolism is not reached for at least four weeks. In three months, the basal metabolic rate falls again to a level averaging 741 calories per square meter, which is maintained for at least five and one-half months.

The values for the later groups of birds do not reveal the expected sex difference, since the pullets showed on an average the greater basal metabolic rate. However, this is partly due to the fact that the area of the combs and wattles have been included in the surface areas used in these computations. Whether this is justifiable or not will depend upon the final explanation for the so-called "surface area" law; that is, whether it is based purely upon the mechanics of heat emission, or upon an anatomical relation between body surface and active protoplasmic material. If the areas of these appendages are not considered, the averages become 742 and 776 calories per square meter for the cockerels and 788 and 761 for the pullets.

TABLE 4.—*The basal heat production of White Plymouth Rock cockerels*

Date	Bird No.	Age	Weight	Rump to shoulder	Surface area	Maximum and minimum temperature of chamber	Work record	Oxygen consumed daily	Respiratory quotient	Heat produced daily		
										Total	Per kilogram body weight	Per square meter body surface
1923		*Days*	*Gm.*	*Cm.*	*Sq. cm.*	*° F.*		*Liters*		*Cals.*	*Cals.*	*Cals.*
Apr. 30	2082	35	278	.....	320	73–87	.....	9.4	0.81	45	164	1,419
May 1	2307	36	275	.....	316	65–86	.....	10.7	.71	51	184	1,601
May 3	2617	38	304	.....	350	84	.....	9.3	.89	46	151	1,314
May 4	2637	39	276	.....	318	62–84	.....	9.4	.80	46	165	1,431
Average	.....	37	283	.....	326	.....	.....	9.8	.80	47	166	1,441
June 6	2097	72	738	16.1	843	76–85	.....	13.7	.77	65	88	770
June 7	2617	73	668	14.6	698	74–84	.....	12.7	.70	60	105	860
June 9	2082	75	628	14.6	734	72–77	.....	15.0	.70	70	112	960
June 11	2307	77	695	15.5	800	72–84	.....	14.8	*.60	70	100	872
June 12	2637	78	640	14.3	732	74–86	.....	11.6	*.68	55	85	744
June 13	2885	79	827	16.5	890	78–90	.....	14.9	.70	70	85	780
Average	.....	76	683	.....	783	.....	.....	13.8	.69	65	96	832
July 24	2097	120	1,306	19.4	1,297	78–90	.....	25.7	*.66	121	87	936
July 25	2885	121	1,443	19.4	1,319	74–83	.....	26.9	.71	126	88	958
July 26	2307	122	1,074	18.3	1,099	76–84	.....	18.0	.71	84	79	768
July 27	2308	123	1,504	18.8	1,321	78–83	.....	24.2	.73	114	76	864
July 28	2009	124	1,201	18.5	1,169	75–82	.....	19.8	.71	93	77	794
Average	.....	122	1,324	.....	1,241	.....	.....	23.0	.70	108	81	864
Sept. 24	2637	182	1,923	20.7	1,583	83–88	.....	34.0	*.69	161	84	1,017
Sept. 25	2307	183	1,750	21.0	1,523	74–87	.....	24.5	*.63	115	66	762
Sept. 26	2097	184	1,980	22.4	1,684	74–90	.....	28.1	*.66	132	66	790
Sept. 27	2308	185	2,225	21.4	1,737	73–81	.....	33.3	*.68	157	71	904
Sept. 29	2009	187	1,761	20.4	1,502	68–81	.....	26.2	*.69	123	70	821
Average	.....	184	1,928	.....	1,606	.....	.....	29.3	.67	138	71	859
Nov. 20	2308	239	3,025	22.1	2,065	68–86	13	42.6	.72	200	66	969
Nov. 21	2307	240	2,065	22.5	1,725	70–96	12	30.0	.72	141	68	817
Nov. 22	2097	241	2,792	23.6	2,063	71–90	8	37.2	.72	174	62	843
Nov. 23	2009	242	2,609	22.8	1,954	72–84	11	35.7	*.69	170	65	870
Nov. 24	2637	243	2,941	23.0	2,085	68–80	13	37.0	*.67	176	60	844
Dec. 1	2905	250	2,797	22.0	1,980	75–88	13	35.9	.71	166	59	838
Average	.....	242	2,705	.....	1,979	.....	.....	36.4	.70	171	63	864
1924												
Feb. 25	2862	336	2,322	20.3	1,719	80–94	17	36.3	*.69	171	74	995
Feb. 26	2097	337	2,490	23.3	1,934	54–90	6	29.7	.72	140	56	724
Feb. 29	2905	340	2,942	21.9	2,025	80–90	8	43.1	.73	203	69	1,002
Mar. 1	2009	341	2,764	23.4	2,043	78–92	5	43.1	.73	203	73	994
Mar. 2	2308	342	2,914	23.0	2,076	82–88	8	33.6	.71	158	54	761
Mar. 3	2637	343	2,934	23.0	2,083	78–86	3	29.4	.72	138	47	663
Average	.....	340	2,728	.....	1,980	.....	.....	35.9	.72	169	62	856

* In estimating the heat production for periods in which the observed respiratory quotient was less than 0.707, the latter respiratory quotient was assumed.

The last group of experiments to be considered on the relation between basal heat production and age and sex includes the first experiments undertaken on the gaseous exchange of chickens. In some respects the conditions under which these experiments were performed were not entirely satisfactory. However, they constitute the most complete set of experiments that has been carried out, and the results appear to be of great significance.

The chickens used were White Plymouth Rocks. At approximately monthly intervals these were removed from the range at the university poultry farm, brought to the laboratory in groups of five or six of like sex, and after a fast of 48 hours (except the first group of

cockerels, which were fasted for approximately 24 hours only), they were placed in a cylindrical animal chamber of appropriate size and subjected to a respiration test of 22 to 23 hours' duration. In most of the tests the activity of the birds during the period of observation was neither controlled nor observed, but the close quarters in which they were confined, combined with the complete darkness which prevailed within the animal chamber, appeared to have effectively discouraged any considerable muscular movements. The temperature of the laboratory, unfortunately, varied widely during these tests, as indicated by the records of a maximum and minimum thermometer placed within the animal chamber, and during the coldest weather it may be suspected that the heat production of the birds was raised to some extent above the basal.

TABLE 5.—*The basal heat production of White Plymouth Rock pullets*

Date	Bird No.	Age	Weight	Rump to shoulder	Surface area	Maximum and minimum temperature of chamber	Work record	Oxygen consumed daily	Respiration quotient	Heat produced daily		
										Total	Per kilogram body weight	Per square meter body surface
1923		*Days*	*Gm.*	*Cm.*	*Sq. cm.*	*° F.*		*Liters*		*Cals.*	*Cals.*	*Cals.*
June 25	2458	91	668	15.2	775	81–86		11.1	0.76	53	79	684
June 26	2070	92	613	14.0	707	71–86		11.9	.74	56	92	800
June 27	2466	93	827	16.0	890	66–72		14.6	*.69	69	83	771
June 28	2441	94	779	16.9	892	67–72		16.3	.70	77	98	861
June 29	2581	95	684	15.3	788	70–76		12.6	.71	59	87	751
June 30	2191	96	821	16.3	896			15.5	*.68	73	89	816
July 2	2281	98	750		815	75–82		14.4	*.65	68	91	834
Average		94	735		824			13.8	.70	65	88	788
July 30	2441	126	1,069	17.4	1,063	78–84		16.7	.75	80	74	749
July 31	2070	127	839	16.1	809	80–87		11.9	.70	56	67	691
Aug. 1	2458	128	1,017	17.5	1,041	82–87		16.7	.70	78	77	753
Aug. 2	2466	129	1,022	16.5	1,007	80–86		16.3	.71	77	75	763
Aug. 3	2191	130	1,138	17.6	1,105	81–92		19.3	*.61	91	80	824
Aug. 4	2791	131	1,169	18.6	1,157	81–86		19.2	.71	90	78	782
Average		128	1,042		1,030			16.7	.70	79	75	760
Oct. 1	[b]2191	189	1,597	19.8	1,404	70–76		27.0	*.68	127	80	907
Oct. 2	2458	190	1,392	18.9	1,275	69–74		21.9	.70	99	71	780
Oct. 3	2686	191	1,455	18.5	1,287	66–74		23.9	.71	112	77	871
Oct. 4	2070	192	1,006	15.9	977	64–73		13.9	.71	65	65	667
Oct. 5	2441	193	1,600	18.8	1,363	60–72		28.9	.70	135	85	994
Oct. 6	2791	194	1,651	20.0	1,437	57–66		28.9	.72	136	82	948
Average		192	1,450		1,290			24.1	.70	113	77	861
Nov. 28	2333	247	1,834	19.2	1,476	64–84	8	25.8	.70	121	66	819
Nov. 30	2441	249	2,177	18.8	1,590	48–82	6	29.9	.72	141	65	885
Dec. 2	2087	251	2,162	20.8	1,683	66–84	5	28.4	*.63	134	62	801
Dec. 3	2376	252	1,777	19.0	1,446	77–84	13	22.4	.61	106	60	737
Dec. 4	2686	253	1,716	19.0	1,420	74–82	3	20.6	.72	97	57	682
Dec. 5	2791	(254)	(2,066)	19.9	(1,602)	72–82	34	(34.1)	(.72)	(161)	(78)	(1,002)
Average		251	1,933		1,523			25.4	.68	120	62	785
1924												
Mar. 7	2866	347	1,677	19.3	1,417	80–88		25.7	.71	121	72	852
Mar. 13	2376	353	2,375	19.9	1,718	75–85	4	25.9	.71	121	51	707
Mar. 14	2333	354	1,906	18.1	1,454	74–86	3	28.3	.72	133	70	914
Do	2441	354	2,378	19.8	1,714		2	31.0	.72	146	61	852
Mar. 18	2694	358	2,265	20.5	1,708	82–94	4	26.7	.72	125	55	733
Mar. 23	2154	363	2,504	21.2	1,832	78–84	7	27.9	.78	131	52	716
Average		355	2,184		1,640			27.6	.73	129	60	796

* In estimating the heat production for periods in which the observed respiration quotient was less than 0.707, the latter respiration quotient was assumed.

[b] This bird was fasted for 29 hours only.

If the birds originally selected for this work died or were used for other purposes, they were replaced by others of the same breed and sex. The experimental observations summarized in Tables 4, 5, and 6 refer to groups of cockerels, pullets, and capons.

TABLE 6.—*The basal heat production of White Plymouth Rock capons*

Date	Bird No.	Age	Weight	Rump to shoulder	Surface area	Maximum and minimum temperature of chamber	Work record	Oxygen consumed daily	Respiratory quotient	Heat produced daily		
										Total	Per kilogram body weight	Per square meter body surface
1923		*Days*	*Gm.*	*Cm.*	*Sq. cm.*	*° F.*		*Liters*		*Cals.*	*Cals.*	*Cals.*
July 3	2085	99	870	16.0	912	77–81	.....	15.3	*0.61	72	83	788
July 6	2207	102	916	16.4	950	83–91	.....	16.4	* .64	77	84	814
July 7	2870	103	1,020	17.2	1,032	82–90	.....	19.2	.74	91	90	878
July 8	2365	104	936	17.3	992	80–90	.....	16.2	.78	77	83	780
Average		102	935	.....	971	.....	.....	16.8	.69	79	85	815
Aug. 6	2207	133	1,258	17.2	1,146	80–94	.....	18.0	.73	85	67	739
Aug. 7	2870	134	1,324	18.6	1,232	79–87	.....	20.5	*.67	96	73	782
Aug. 8	2085	135	1,239	17.7	1,157	78–93	.....	20.0	.70	94	76	812
Aug. 9	2365	136	1,229	18.7	1,190	82–93	.....	16.5	*.67	78	63	652
Aug. 10	2364	137	1,847	19.9	1,515	85–94	.....	31.3	.72	147	80	970
Aug. 11	2022	138	1,152	19.6	1,186	85–92	.....	21.4	.72	100	87	847
Average		135	1,341	.....	1,238	.....	.....	21.3	.70	100	74	800
Oct. 9	2022	197	1,296	20.7	1,299	70–80	.....	22.8	.73	108	83	828
Oct. 10	2365	198	1,934	19.9	1,550	72–80	.....	16.5	.74	78	40	501
Oct. 11	2085	199	1,306	20.5	1,297	73–80	.....	20.1	.71	95	72	730
Oct. 12	2364	200	2,517	22.5	1,904	71–78	.....	37.7	.72	178	71	933
Average		199	1,763	.....	1,512	.....	.....	24.3	.72	114	66	748
Dec. 10	2364	259	3,297	23.2	2,219	62–88	3	46.1	.72	213	65	960
Dec. 11	2365	260	2,234	20.1	1,676	62–88	3	19.7	.72	92	41	551
Dec. 12	2022	261	1,629	21.1	1,474	74–78	.....	23.2	*.62	109	67	739
Dec. 14	2695	263	2,493	23.0	1,920	68–82	13	29.7	.71	139	56	724
Dec. 15	2085	264	1,509	19.8	1,365	72–94	7	19.7	.70	92	61	677
Dec. 19	2132	268	2,437	23.3	1,913	80–84	24	31.6	.71	148	61	772
Average		262	2,266	.....	1,761	.....	.....	28.3	.70	133	59	737
1924												
Mar. 24	2733	364	2,610	21.7	1,897	72–86	1	30.3	.70	142	54	748
Mar. 25	2132	365	3,105	22.7	2,126	68–80	9	29.7	*.68	140	45	658
Mar. 26	2272	(366)	(2,277)	.....	(1,705)	78–98	17	(33.0)	(.74)	(156)	(68)	(915)
Mar. 27	2725	367	2,935	23.2	2,101	78–85	7	37.7	.71	176	60	840
Mar. 28	2364	368	3,864	23.8	2,440	81–86	4	45.4	.73	213	55	874
Mar. 29	2412	369	3,379	21.3	2,135	79–84	5	34.2	.73	161	48	756
Average		366	3,183	.....	2,140	.....	.....	35.5	.71	166	52	775

* In estimating the heat production for periods in which the observed respiratory quotient was less than 0.707, the latter respiratory quotient was assumed.

The last-named were caponized at 70 days of age. The surface areas of these birds, except those of the youngest group, were estimated from their body weights and their measurements from the shoulder to the rump, according to the formula (*30*) $S = 5.86\ W^{0.5}\ L^{0.6}$, in which S is the surface area in square centimeters, W the body weight in grams, and L the rump-to-shoulder distance in centimeters. The areas of the youngest group of cockerels were estimated from the determined areas of five 0.5-pound birds, on the assumption that the area varied as the two-thirds power of the body weight.

For convenience in studying the large numbers of data in these tables, the group averages have been summarized in Table 7.

TABLE 7.—*A summary of the average results in Tables 4, 5, and 6*

Number of birds	Age	Weight	Surface area	Heat produced daily		
				Total	Per kilogram body weight	Per square meter body surface
	Days	*Gm.*	*Sq. cm.*	*Cals.*	*Cals.*	*Cals.*
COCKERELS						
4	37	283	326	47	166	1,441
6	76	683	783	65	96	832
5	122	1,324	1,241	108	81	864
5	184	1,928	1,606	138	71	859
6	242	2,705	1,979	171	63	864
6	340	2,728	1,980	169	62	856
PULLETS						
7	94	735	824	65	88	788
6	128	1,042	1,030	79	75	760
6	192	1,450	1,290	113	77	861
5	251	1,933	1,523	120	62	785
6	355	2,184	1,640	129	60	796
CAPONS						
4	102	935	971	79	85	815
6	135	1,341	1,238	100	74	800
4	199	1,763	1,512	114	66	748
6	262	2,266	1,761	133	59	737
5	366	3,183	2,140	166	52	775

The first group of cockerels, averaging 37 days of age, showed a relatively intense basal heat production. Apparently these birds were near, if not at, the peak of rising basal metabolism (per unit of surface) noted in the preceding studies on young chicks. Between the age of this group and the age of the next group (76 days) the intensity of the basal metabolism apparently suffered a rapid decline. In this connection some results on a group of six Rhode Island Red chicks, averaging 55 days of age, may be considered. (See Table 8.)

TABLE 8.—*The basal heat production of a group of young Rhode Island Red chicks*

Date	Bird No.	Age	Weight	Surface area	Maximum and minimum temperature of chamber	Oxygen consumed daily	Respiratory quotient	Heat produced daily		
								Total	Per kilogram body weight	Per square meter body surface
1923		*Days*	*Gm.*	*Sq. cm.*	*° F.*	*Liters*		*Cals.*	*Cals.*	*Cals.*
June 18	C429	53	240	345	81–88	6.48	0.70	30.4	126	881
June 19	C385	54	238	340	80–89	6.61	.69	31.0	130	912
June 20	C485	55	223	325	82–91	6.68	[a].70	31.3	140	953
June 21	C383	56	200	300	82–92	5.90	.70	27.6	137	920
June 22	C491	57	283	390	83–93	7.64	[a].70	35.8	127	918
June 23	C...	58	250	360	.....	7.16	.69	33.4	134	928
Average	.....	55	239	343	.....	6.74	.70	31.6	132	920

[a] Assumed. The observed respiratory quotient was considerably smaller.

The experiments on these chicks were conducted at about the same time as the other experiments, and exactly the same procedure was followed. The sex of these birds was not recorded. The average basal heat production for the group was 920 calories per square meter of body surface (estimated from skin area determinations of White Plymouth Rock birds of approximately the same size), the individual determinations being remarkably concordant. These results, in comparison with those obtained on the two youngest groups of White Plymouth Rock cockerels, indicate that the intensity of metabolism which had reached its peak at 30 to 40 days of age was greatly reduced at 55 days, though still distinctly higher than the level observed three weeks later.

It is a matter of considerable significance that, for the cockerels and pullets, the basal heat production per square meter of body surface was remarkably constant for all age groups, except the youngest cockerels, while the basal heat production per kilogram of body weight decreased continuously with advancing age. This fact constitutes a formidable argument in favor of the body surface as a unit of reference for basal metabolism as opposed to the body weight.

Whatever the ultimate explanation of the relation between basal heat output and body surface may prove to be, it is undoubtedly the closest relation involving a factor of body size that has thus far been discovered. It has been pointed out previously (*13*) that it is contrary to fact to assume, as is frequently done in explaining the surface area law, that the active protoplasmic tissue varies in proportion to the body surface.[4] On the other hand, it appears reasonable to suppose that in the evolution of species the intensity of the basal heat production should be adjusted to such a level that, at temperatures so extreme as to threaten the continued life of the animal, the physiological mechanism for restricting heat emission at low temperatures or increasing it at high temperatures should be equally effective among animals differing in size. This would be true if the basal heat production per unit of surface were approximately the same for all (warm blooded) animals, since the extent of surface is the main anatomical factor determining heat emission, either by radiation, the method of heat loss prevailing at low temperatures, or by the evaporation of water, the only method operative at environmental temperatures above that of the animal body itself.

In previous experiments (*28*) with mature Rhode Island Red cocks and hens, average values of 806 and 703 calories were obtained. The same method was used and much the same conditions prevailed as in the work with the White Plymouth Rocks. Whether these lower values, especially for female birds, are due to the greater age of the birds, to a lesser activity, or to a more prolonged confinement in the laboratory can not at present be determined. In the hope of throwing some light upon this point, basal metabolism studies were made with four mature White Plymouth Rock hens. The periods of observation were short (30 to 50 minutes) and were taken when the hen was completely at rest. Only the CO_2 production was deter-

[4] The same situation exists in comparing man with the chicken as in comparing man with the rat. Thus, a man of average height (173 cm.) and weight (70 kgm.) possesses a surface area (1.83 sq. m.) nine and two-tenths times as great as the surface area of a White Leghorn cockerel weighing 2 kgm. If the protoplasmic matter in animals varies with the body surface, the man should contain only 18 kgm., or 26 per cent, of protoplasm, on the extreme assumption that the cockerel consists entirely of active cellular tissue.

mined; in the computation of the heat production, a basal respiratory quotient of 0.73 was assumed. Several determinations were made upon each hen. The results are summarized in Table 9.

TABLE 9.—*The resting metabolism of mature White Plymouth Rock hens*

Hen No.	Body weight	Estimated body surface [a]	Temperature of chamber	CO_2 per hour				Heat produced daily	
				1	2	3	Average	Total	Per square meter body surface
	Grams	*Sq. cm.*	*°F.*	*Grams*	*Grams*	*Grams*	*Grams*	*Calories*	*Calories*
35	2,290	1,757	78–86	1.52	1.42	--------	1.47	116	661
488	2,900	1,961	68–82	1.48	1.31	1.37	1.39	109	558
50	2,870	2,006	78–84	1.51	1.62	1.53	1.55	123	611
48	3,000	2,011	82–84	1.84	1.78	1.60	1.74	137	684

[a] The surface area was estimated from the body weight and the rump-to-shoulder dimension.

Quite possibly the difference between the heat production per square meter of surface for these hens (averaging 628 calories) and the average obtained for the mature Rhode Island Red hens (703 calories), if significant, is due to a less degree of activity. In any case, it appears probable that the values obtained for the young White Plymouth Rocks (852 calories for cockerels and 805 calories for pullets) are subject to a considerable decrease at a later age.

Comparing the average basal metabolism per unit of surface for the different groups of cockerels with those for the groups of pullets of approximately the same age, it appears that in four of the five possible comparisons the average for the cockerels exceeds that for the pullets. The opposite relation for the groups with average ages of 184 and 192 days may quite probably be due to the low temperatures prevailing during part of the day throughout the observations on the pullets. (See Table 5.) This may have raised the heat production of the pullets of that group above the actual basal level. The average value for all groups of cockerels but the first was 852 ± 12 calories per day per square meter of surface, and that for the pullets was 805 ± 10 calories. The average difference is 47 ± 16 calories. The basal metabolism of the pullets thus averaged 5.5 per cent less per unit of surface than the basal metabolism of the cockerels.

The coefficient of variation of the 28 determinations of the basal metabolism per unit of surface for the cockerels was 11.3 per cent, and for the pullets 10.1 per cent. These coefficients may be compared with those reported by Harris and Benedict (*19*) for men and women, i. e., 8.05 and 9.17, respectively; with those reported by Mitchell and Carman for rats (*27*), i. e., 13.5 for males and 8.0 for females; and with those observed by Mitchell and Haines (*28*) for mature Rhode Island Red chickens, i. e., 10.9 for cocks and 11.6 for hens. Thus, although the experimental conditions in the work on the White Plymouth Rock chickens were not under as good control as would be desirable, still the varying conditions of temperature and activity apparently did not induce an abnormally great variability in the results secured. It is fair to conclude, therefore, that the average

basal heat productions obtained were not seriously vitiated by such uncontrolled factors.

The calorimetric results obtained upon the capons have a bearing upon the question of the effect of castration upon metabolism. Although many reported experiments (*1*, *22*, *23*, *24*), particularly those of Heymans (*22*) on cocks, afford evidence in favor of the view that male castration lowers the basal metabolism, other experimental observations are on record (*2*, *12*, *26*) in which no evidence of such an effect appears. The writers' results speak unmistakably in favor of the view that removal of the testes lowers the basal metabolism. From Table 7, it is evident that the group averages for the capons decreased progressively with increasing time after castration (which occurred at 70 days of age), except for the last group. For seven of the capons, more than one observation was made. These have been summarized for each bird separately in Table 10.

TABLE 10.—*Effect of castration on basal metabolism*

Capon No.	Age	Basal metabolism per square meter body surface	Capon No.	Age	Basal metabolism per square meter body surface
	Days	*Calories*		*Days*	*Calories*
2085	99	788	2365	104	780
	135	812		136	652
	199	730		198	501
	264	677		260	551
2207	102	814	2364	137	970
	133	739		200	933
2870	103	878		259	960
	134	782		368	874
2132	268	772	2022	138	847
	365	658		197	828
				262	739

It is evident that for each capon, with the exception of No. 2364, a marked lowering of metabolism occurred, the decrease extending over several months. No such decrease in basal metabolism was noted with the cockerels or the pullets. Against the possible explanation that the decrease was due to a lowering of muscular activity, it may be pointed out that in the later experiments in which activity records were secured, the indicated activity of the capons (Table 6) was certainly no less than the indicated activity of the cockerels (Table 4) or of the pullets (Table 5). The lowered metabolism seems to be a true effect of castration.

The average basal metabolism of the capons was 779 ± 15 calories per day per square meter of surface, and the average difference between cockerels and capons in this value was 73 ± 19. This appears to be a significant difference, statistically. Actually, the difference between cockerels and capons was greater than this, since the lowering of metabolism induced by castration was not immediately initiated, and since it extended over several months. The maximum effect was probably measured in the group of capons averaging 262 days of age. The average value in this group was 737 calories, as compared with the average of 852 calories for the cockerels, a reduction of 13.5 per cent.

SUMMARY

The effect of age on the basal heat production of chickens has been studied, using several different breeds of birds. A remarkable similarity was observed between chickens and human subjects, in that the metabolism per unit of surface is distinctly below the adult level at hatching (birth), rises rapidly to a maximum, and then decreases again to an adult level that is maintained for a considerable fraction of the life span. For the chicken, the peak in the curve appears to be reached at 30 to 40 days of age, and the adult level at about 70 to 80 days.

The basal metabolism of chickens per unit of weight decreases continuously from the peak age to 1 year of age at least. For this species of animals the greater constancy of the basal heat production, when referred to surface than when referred to weight, is distinct and marked.

The basal metabolism of cockerels is eventually higher than that of pullets, though the exact age at which the sex difference appears has not been determined. In a group of White Plymouth Rock chickens ranging in age from 72 to 363 days, the average basal metabolism of the cockerels was 852 ± 12 calories per day per square meter of surface, and that of the pullets was 805 ± 10 calories. At more advanced ages there appeared to be a further reduction in basal heat production of 6 to 12 per cent for cocks and hens, respectively.

The castration of cockerels lowers their basal metabolism, though the effect may not be immediate and, when initiated, may extend over several months. The average maximum effect observed was a reduction of 13.5 per cent.

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THE INHERITANCE OF OVATE AND RELATED SHAPES OF TOMATO FRUITS[1]

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INTRODUCTION

Ovate or elliptical shapes of tomato fruits, characteristic of such commercial varieties as the Red or Yellow Plum and similar types, including the Red or Yellow Pear varieties, occupy an intermediate position in the matter of inheritance. They are not inherited as distinctly as are the color characters of the fruit or certain morphological characters (dwarf, peach, leaf types, inflorescence types), nor are they as complex genetically or as susceptible to environmental agencies as the more quantitative character of fruit size. From a genetic standpoint, they may be termed semiquantitative in nature.

Circumstantial evidence, verging on complete proof, has indicated that the inheritance of ovate shape in tomato fruits may conveniently be explained as being controlled largely by a single, major gene carried in the first linkage group of Lycopersicum (*3*).[3] Experimental data are now available to confirm this, and to afford the necessary quantitative demonstration of the situation.

METHODS

Perhaps the most direct, accurate, and concise description of shape in tomato fruits is given by the ratio of the measured equatorial diameter of the fruit (taken midway between the stem and blossom end) to the polar diameter, taken at right angles to the equatorial. This ratio may be expressed as an index number. Accordingly, in this report the term "shape index" is defined as the following ratio:

$$\text{Shape index} = \frac{\text{equatorial diameter}}{\text{polar diameter}}$$

With such an index, a round fruit will be rated at approximately 1.00, an ovate type about 0.75, and an oblate fruit will vary from 1.15 upwards. Shape measurements were made with small steel calipers, readings being taken to the nearest millimeter.

Up to the present the writer has not attempted to correct the polar diameter measurements in fruits possessing a deep cavity or basin at the stem end. With such fruits care was taken to place the calipers in such a manner as to give an average reading for the ofttimes irregular surface of the fruit. Accordingly, the shape indices of the larger oblate fruits with deep cavities will be smaller than if the

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[2] M. R. Irwin, graduate student in genetics, has given valuable assistance in the large series of fruit measurements involved in these investigations.

[3] Reference is made by number (italic) to "Literature cited," p. 985.

measurement had been taken at the stem scar. This minimization of the shape index of the larger fruits is not considered as seriously affecting the general shape relationships. It merely affects the frequency distribution or curve at the upper end a trifle, and the results are probably a bit conservative if differences are being considered as they are in this report.

Ordinarily eight or more tomato fruits from a single plant are measured. In some earlier work only five fruits were used, but this number has now been increased so that 10 are used if possible, since this simplifies the calculation of averages. The tomato plants are trained to a single stem in the greenhouse, and the fruits to be measured come from the first four or five clusters. In the field very little pruning is practiced until the first five clusters have set, after which the tops are pruned rather severely.

In the field the plants of a series are replicated in short rows, 10 plants to the row. In the greenhouse a lot of plants (usually 200 to 400) are grown in 8-inch pots, and are shifted five or six times to various places on the greenhouse benches so as to equalize growing conditions as much as possible.

EXPERIMENTAL DATA

The inheritance of the ovate fruit shape will be considered first, followed by the genetic relations of this shape factor to other factors, such as dwarf, peach, and fruit size. In a later section the mode of inheritance of the oblate and round fruit shapes will be presented, primarily for comparison with the behavior of ovate shape.

INHERITANCE OF OVATE FRUIT SHAPE

The commercial varieties (with the abbreviations) involved in the study of ovate fruit shape were as follows:

Oblate types	*Ovate types*	*Round type*
New Globe (NG).	Yellow Plum (YP).	Red Peach (RP).
Dwarf Stone (DS).	Red Pear (RPr).	

These varieties have been crossed under controlled conditions, and F_1, F_2, and back-cross generations have supplied the material for the measurements.

The elliptical or ovate shape was introduced into the hybridization tests by the Yellow Plum variety. Two oblate and one round or roundish-oblate variety served to give the contrasting character in the three crosses that are listed in Table 1, which gives the shape indices of the P_1, F_1, and F_2 generations. In all cases each of these three generations has been grown and compared during the same season and under similar conditions.

TABLE 1.—*Mean fruit shape indices of three crosses of ovate by oblate varieties*

Crosses	Ovate parent	Oblate parent	F_1	F_2	Where and when grown
Yellow Plum×New Globe	0.71	1.22	1.08	1.06	Field, 1926.
Yellow Plum×Dwarf Stone	.75	1.34	1.15	1.13	Greenhouse, 1926.
Yellow Plum×Red Peach	.71	1.15	.98	.97	Field, 1926.

As measured by the shape indices, the first hybrid generation shows an intermediate condition in all the crosses. The fruits tend to be more or less round. In all cases, however, there is a slight dominance of the oblate shape. The F_2 generations exhibit an average condition approaching the F_1 shape, but naturally this generation is highly variable, as may be noted in the subsequent graphs (fig. 1). Both parental shapes are usually recovered in this F_2 generation, a situation which ordinarily does not occur when fruit sizes are involved in tomato hybrids. The F_2 data of the three frequency distributions are plotted in Figure 1, the solid line in the graphs representing the total F_2 frequency.

Whereas all three curves in this figure approach a normal curve, there is sufficient evidence of multimodality to occasion some consideration of these F_2 distributions. To one with a predilection for a simple, monohybrid explanation of the case, there is a hint of trimodality in these curves, suggestive of a 1:2:1 distribution. The secondary modes on both sides of the major modes may be explained away as modifying factors of some sort. To the skeptic, however, the curves probably exemplify the capricious fluctuations of such characters as shape. At any rate there would be serious difficulty in attempting to define the limits of the three genotypes in a monohybrid population. Apparently F_2 distributions of shape characters are uncertain measures of hereditary genotypes, whether caused by environmental influences or germinal complexity. Accordingly, one must turn to other devices to interpret the case.

The solution of the difficulties noted above proceeded from a consideration of the back cross of the F_1 of an ovate-oblate cross to the ovate parent. Several F_1 plants of the Yellow Plum-Dwarf Stone cross were reciprocally crossed by the Yellow Plum type. They were also crossed with the unrelated, ovate (pyriform) type, the Red Pear.

Shown in Figure 2 are the curves of the two back crosses. In the Yellow Plum back-cross generation of 276 plants there is exhibited a bimodal curve of unusual distinctness. The conclusion is evident that two major factors are acting with pronounced effect, there being no evidence of secondary or minor influences beyond the fluctuations to be expected in such characters as shape. Presumably two hereditary genotypes are responsible for the two modes at the classes 0.80 and 1.10. This pointed suggestion, coupled with the hint of trimodality in the F_2 generations, leads to the reasonable deduction that ovate shape in tomato fruits, when allelomorphic to the oblate shape, may very well be governed by a single, major gene. The back-cross condition of bimodality would then represent the action of the heterozygous and the homozygous recessive genotypes.

Verification for such a deduction is afforded by the cross of similar F_1 plants (Yellow Plum × Dwarf Stone) to the ovate (and pyriform) Red Pear variety. Such a cross involving 89 plants is shown graphically in the upper part of Figure 2. Again an equally distinct, bimodal curve results, giving evidence that the pyriform shape of the Red Pear variety is genetically equivalent to the ovate shape of the Yellow Plum as far as the ellipsoidal features of its shape are concerned.

It is probable that pyriform and true ovate shape differ only in a secondary manner. Ovate shape seems to be incompletely domi-

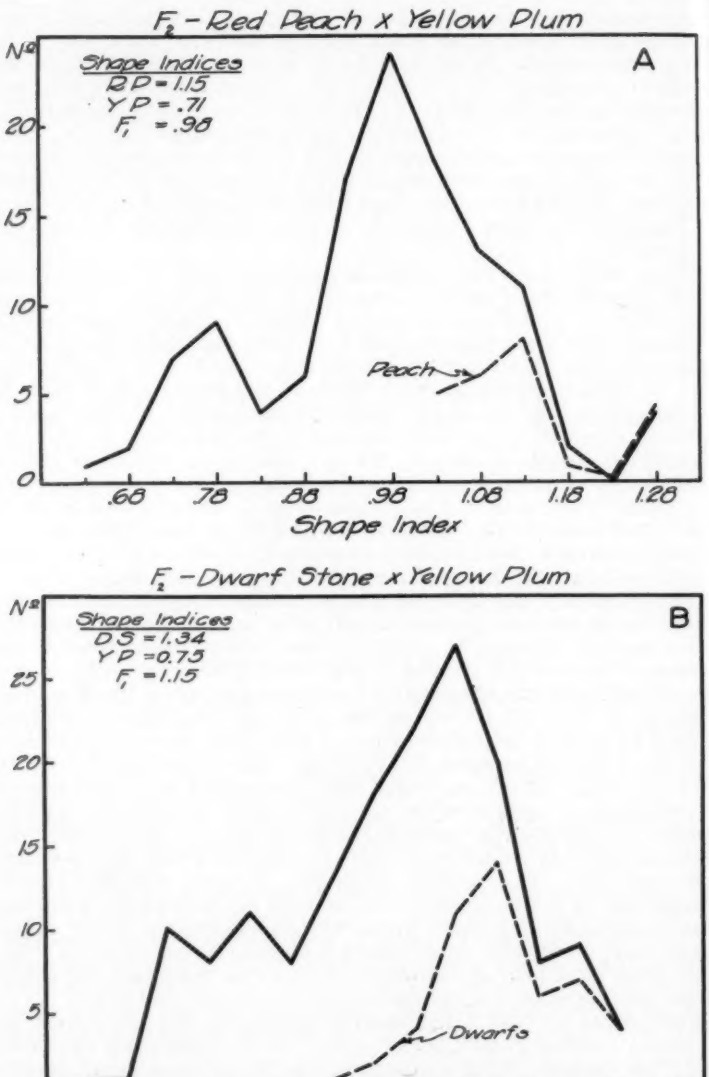

FIG. 1.—Frequency distributions of three different F_2 generations from crosses of ovate×oblate varieties, showing tendency for trimodality in fruit shape; solid line represents total F_2 frequency

nant over pyriform shape. Very often in crosses of oblate×ovate (nonpyriform), a few pear-shaped types appear, despite the fact that both parents were homozygous for their respective fruit shapes. In the New Globe×Yellow Plum cross discussed herein, for example, the following proportion of pear fruits was observed: 99 oblate or round; 24 ovate (nonconstricted); 12 pyriform (0 distinct, 5 medium, 7 slight pear-shaped). This seems to indicate that oblate varieties may sometimes carry pyriform shape as a cryptomere, since there is other evidence to prove that the homozygous Plum varieties are not responsible for the pear shape.

This suggestion is confirmed in the back cross of the F_1 of the Dwarf Stone-Yellow Plum cross to the Red Pear variety, in which the following distribution of shapes occurred: 41 oblate and round; 24 ovate (nonpyriform); 9 slight pyriform; 13 medium pyriform; 2 distinct pyriform. Apparently one half of the elliptical fruits showed

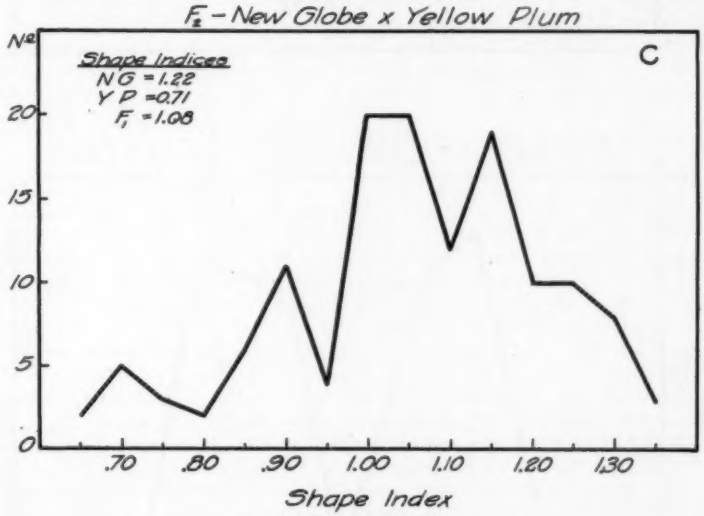

FIGURE 1—Continued

some constriction of the neck (pyriform), the other half not. This suggests that the ovate gametes from the F_1 of an oblate×ovate cross (such as Dwarf Stone×Yellow Plum) are of two sorts, one with the true ovate gene coming from the P_1 Yellow Plum variety, and the other with a pyriform gene which probably was contributed by the oblate Dwarf Stone variety.

It is also worth noting that in some crosses of oblate by pyriform types the pear shape certainly does not appear in F_2 in its full quota of 25 per cent, as might be expected from the results of MacArthur (*5*). There is always a marked deficiency of true pear-shaped fruits in F_2. For example, in two crosses for which only descriptions of the fruit shapes are available the proportion of pyriform types was as follows:

F_2—Ponderosa×Red Pear—28 nonpyriform; 3 pyriform.
F_2—Dwarf Peach×Red Pear—190 nonpyriform; 20 pyriform (including 4 distinct, 7 medium, and 9 slightly constricted types).

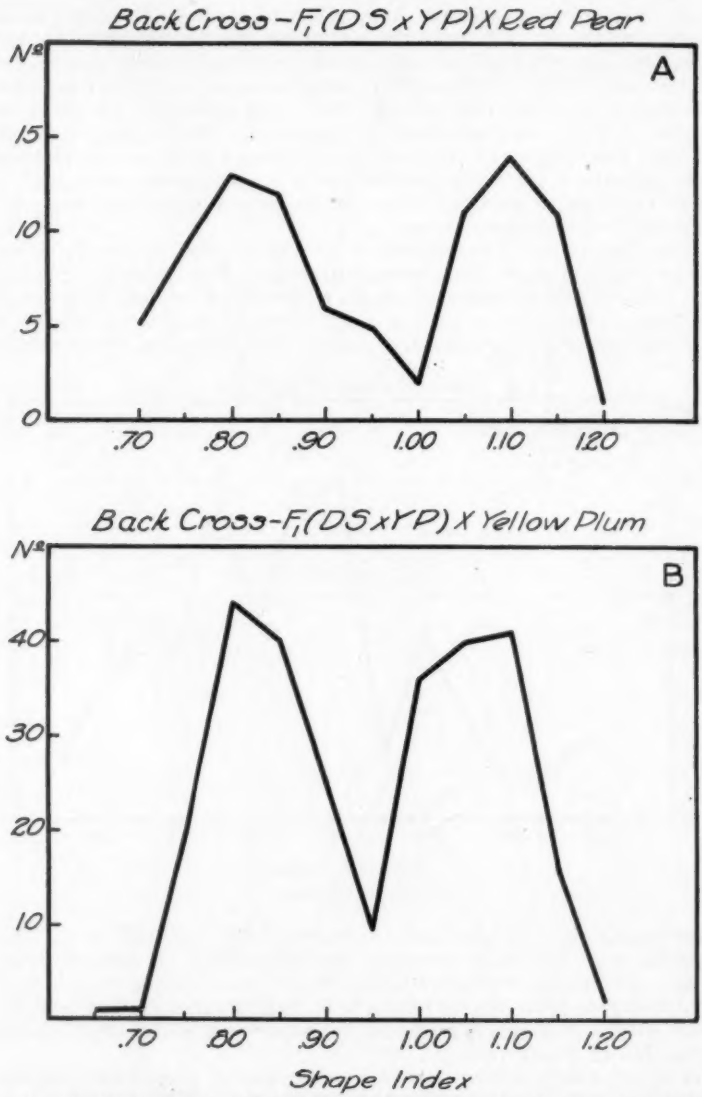

FIG. 2.—Two back-cross generations showing distinct bimodality in fruit shape. F_1 plants of the Dwarf Stone×Yellow Plum cross were crossed to the ovate Yellow Plum (below) and also to the pyriform Red Pear (above). The plants were grown in the greenhouse at the same time as the F_2 generation in Figure 1

Among the nonpyriform fruits in these F_2 generations are classed the true ovate or plum types. Were these added to the pyriform types, the combined percentage would approximate the expected 25 per cent. These results confirm the previous findings that ovate and pear shapes are conditioned by the same hereditary factor as far as their their ovate shape is concerned; in which case it is presumably not the pyriform gene but the ovate gene that is the simple allelomorph of the oblate shape gene. The factor for pyriform shape accordingly belongs elsewhere, and as far as the writer is aware, its true genetic status is practically unknown. It is possible that MacArthur's (*5*) report on the simple recessive nature of the pear gene differs from the writer's conception of the case by reason of the use of the Yellow Pear variety by MacArthur as the source of his pyriform gene. It is difficult to conceive, however, that the Yellow Pear variety, which is identical in appearance with the Red Pear variety used herein, should carry a different sort of shape gene. The situation needs further work. As far as the writer is concerned, however, the main point has been established, namely, that ovate and pyriform shapes act similarly from a genetic standpoint in crosses with the oblate types, indicating that they possess the same ovate-determining gene.

If this is true it is more than likely that ovate shape in tomato fruits is in general governed by a single Mendelian factor, at least when studied in contrast with the oblate or roundish-oblate fruit shapes. Despite its semiquantitative nature, it can be isolated in back crosses at least, sufficiently delimited to afford some reasonable assurance of its presence. Under such circumstances its exact location in a particular linkage group becomes possible.

The genetic symbol for this ovate gene might be given as *oo*, since this report is the first to demonstrate its existence quantitatively. This symbol was suggested in an earlier paper (*3*), but an abstract by MacArthur (*4*) had suggested the symbol *pr* for the pyriform shape, inferring that this was the gene located in the first linkage group. Since this shape with the constricted neck is now known to be genetically similar to the ovate form, it is probable that the latter is the true shape gene residing in the first chromosome. As the matter now stands the use of the *Oo* symbols as factors for the oblate-ovate relation is the more descriptive and fundamental, and also the use of a biliteral symbol *pr* is likely to prove unwieldly in view of a more complex situation in fruit shape about to be described. In this case, multiple allelomorphs are probably concerned which will necessitate at least a triliteral symbol for each of the fruit shapes concerned. Accordingly, it is more advantageous to use the simplest symbolism, waiving for this reason the doubtful correctness of the *pr* factor symbol.

LINKAGE OF OVATE FRUIT SHAPE AND TALL-DWARF FACTORS

Since it has been demonstrated by a quantitative analysis that ovate fruit shape is probably governed by a single, major gene, it is possible to ascertain more or less exactly the linkage relations of this gene by means of the same quantitative measurements of fruit shape. Earlier work by Lindstrom (*3*) and by MacArthur (*5*) has indicated that the ovate gene belongs in the first linkage group of

the tomato. Accordingly it should exhibit linkage with the tall-dwarf (*Dd*) and smooth-peach (*Pp*) factor pairs.

Experimental data on the linkage between the tall-dwarf and ovate genes are now available from the cross of the Yellow Plum by Dwarf Stone varieties. Both F_2 and back-cross results have been procured in this cross. The back-cross data, being more reliable will be presented first.

TABLE 2.—*Linkage relations between fruit shape and dwarf factors in reciprocal back crosses of the Dwarf Stone×Yellow Plum F_1 by Yellow Plum tested by seedling progenies*

Shape index	F_1×Yellow Plum				Yellow Plum×F_1				Grand total
	DD	*Dd*	Untested	Total	*DD*	*Dd*	Untested	Total	
0.68					1			1	1
0.71					0			0	0
0.74	1			1	2			2	3
0.77	5	1		6	5		1	6	12
0.80	3	1		4	14	2		16	20
0.83	12	2		14	16	1		17	31
0.86	14	3		17	8	3		11	28
0.89	11	0		11	7	1		8	19
0.92	2	0		2	9	2	1	12	14
0.95	4	0		4	2	0		2	6
0.98	1	1		2	3	2		5	7
1.01	2	5		7	2	6		8	15
1.04	3	10	1	14	1	13		14	28
1.07	1	12	1	14	2	11	1	14	28
1.10	2	8	0	10	0	19		19	29
1.13	1	3	1	5	0	13		13	18
1.16	2	7		9	1	3		4	13
1.19		1		1		1		1	2
1.22						2		2	2
Total	64	54	3	121	73	79	3	155	276

In Table 2 are given reciprocal back-crosses of F_1 (DS×YP)× Yellow Plum. Since no double-recessive (ovate-dwarf) type was available at the time this material was grown, it became necessary to use the Yellow Plum (ovate-tall) variety. This requires a further seedling test to determine the segregation of the *Dd* genes. This is easily done, since tall and dwarf types may be determined in the seedling stage without difficulty or uncertainty. In Table 2, accordingly, 270 progenies of seedlings were grown from the back crosses noted above. At least 30 seedlings were used for each progeny; usually the average was 40 or more. In cases of doubt, arising ordinarily because of small or abnormal seedlings which might be confused with dwarfs, two or three plantings of 40 seedlings each were grown. With 40 seedlings as a test for *DD* or *Dd* genotype on the basis of a 3:1 ratio, the probability of mistaking the genotype by the chance nonappearance of a dwarf, is only 0.05 per cent.

Because of the bimodal nature of the shape curve (see fig. 2) it was relatively simple to draw a reasonably sharp line between ovate and nonovate fruit shapes. From an inspection of Table 2, one may note that very few of the ovate fruit types were of the *Dd* genotype, and but few of the nonovate types of the *DD* genotype, these representing the nonparental or crossover combinations. These data have been summarized in Table 4, where one may note that of the 270 back-cross individuals tested in these reciprocal crosses, only 33 were of the nonparental type. This gives a total crossing-over

percentage of 12.2 (or 9.9 per cent crossing over in microsporogenesis and 15.2 per cent in megasporogenesis).

TABLE 3.—*Linkage relations between fruit shape and dwarf factors in back cross of F_1 (Yellow Plum×Dwarf Stone) by Red Pear tested by seedling progenies*

Shape index	*DD*	*Dd*	Total	Shape index	*DD*	*Dd*	Total
0.68	2		2	1.01	0	1	1
0.71	3		3	1.04	0	5	5
0.74	3		3	1.07	2	6	8
0.77	6	2	8	1.10	1	7	8
0.80	8	0	8	1.13		7	7
0.83	6	1	7	1.16		8	8
0.86	8		8	1.19		1	1
0.89	5		5				
0.92	3		3	Total	50	39	89
0.95	1		1	Mean	0.84	1.08	
0.98	2	1	3				

A somewhat lower percentage was obtained in the back cross of the same F_1 plants to the Red Pear variety (Table 3). Here tests were conducted in the same manner as in the preceding back cross (seedling progenies), and from a total of 89 progenies only six were of the crossover combinations, giving 6.7 per cent crossing over.

If these three back-cross generations are totaled, the average percentage crossing over becomes 10.9, there being 39 nonparental combinations in a total of 359 individuals in the experiment (Table 4). Accordingly, one may conclude that the ovate gene resides rather closely to the dwarf gene in the first chromosome.

TABLE 4.—*Summary of data showing intensity of linkage between fruit shape and tall-dwarf factor pairs*

Crosses	Tall		Dwarf		Percentage crossing over
	Nonovate	Ovate	Nonovate	Ovate	
Back crosses:					
F_1 (DS×YP)×YP♂	11	53	47	7	15.2
YP×F_1 (DS×YP)♂	6	67	70	9	9.9
F_1 (DS×YP)×Red Pear♂	3	47	36	3	6.7
Total	20	167	153	19	10.9
F_2 generation:					
Dwarf Stone×Yellow Plum	72	39	49	0	
Expected with 10 per cent crossing over	80.4	39.6	39.6	0.4	

TABLE 5.—*Linkage relations between fruit shape and dwarf factors in F_2 if reciprocal crosses of Yellow Plum×Dwarf Stone*

Shape index	*D*	*d*	Total	Shape index	*D*	*d*	Total
0.77	1		1	1.16	10	3	13
0.80	0		0	1.19	12	8	20
0.83	5		5	1.22	5	5	10
0.86	6		6	1.25	4	11	15
0.89	5		5	1.28	2	5	7
0.92	6		6	1.31	1	2	3
0.95	7		7	1.34	0	6	6
0.98	5		5	1.37	2	3	5
1.01	4		4	1.40		1	1
1.04	6	1	7	1.43		1	1
1.07	8	2	10				
1.10	11	0	11	Total	111	49	160
1.13	11	1	12	Mean	1.08	1.25	1.13

Verification for this linkage is afforded by the F_2 generation of the Dwarf Stone × Yellow Plum cross (Table 5 and fig. 1). This gives the same "repulsion" phase of the linkage, and the summary shows 72 tall, nonovate; 39 tall, ovate; 49 dwarf, nonovate; 0 dwarf, ovate, among 160 plants. This fits a linkage hypothesis with 10 per cent crossing over fairly well. (See Table 4.) The absence of the double recessive class is to be expected, since at least 400 individuals are required with 10 per cent crossing over to give an even chance for this type to appear.

Further verification for this degree of linkage was obtained from this same F_2 generation by an F_3 test. In this case, 34 ovate (with shape index of 0.99 or less) tall F_2 plants were self-fertilized, and seedling progenies grown to determine the ratio of the *DD* and *Dd* genotypes. The results appear in Table 6, where there are 28 *DD* and 6 *Dd* types. With independent inheritance this ratio should be 1:2, respectively. With 10 per cent crossing over the fit of observed and theoretical values becomes perfect.

TABLE 6.—*Showing linkage between fruit shape and tall, dwarf factor pairs in ovate, tall segregates from F_2 of Dwarf Stone × Yellow Plum cross, tested by F_3 seedling progenies*

Shape index	Homozygous tall progenies	Heterozygous progenies	Shape index	Homozygous tall progenies	Heterozygous progenies
0.77	1		0.98	2	2
0.80	0				
0.83	4	1	Total	28	6
0.86	5	1	Expected with 10 per cent crossing over	28	6
0.89	4	1			
0.92	6	0	Expected with no linkage	11	23
0.95	6	1			

Thus from four sources there is good evidence of a linkage between the recessive, ovate gene for fruit shape and the dwarf factor characteristic of the Dwarf Stone variety. The intensity of the linkage may be measured by an average value of 10 or 11 per cent crossing over.

LINKAGE OF THE OVATE AND PEACH GENES

Earlier work (*1*, *3*) having demonstrated that the peach and dwarf characters, each being governed by a single recessive gene, were borne very closely together on the first chromosome of the tomato, it should of course follow that the ovate gene is linked with the peach factor. That this is true may be seen from the F_2 data in Table 7 (also in fig. 1) coming from a cross of the Red Peach (round-oblate) × Yellow Plum varieties.

TABLE 7.—*Linkage relations between fruit shape and peach factors in F_2 generations of the cross Red Peach × Yellow Plum*

Shape index	Field planting			Greenhouse planting		
	P	*p*	Total	*P*	*p*	Total
0.63	1		1			
0.68	2		2	2		2
0.73	7		7	3		3
0.78	9		9	2		2
0.83	4		4	6		6
0.88	6		6	1		1
0.93	17		17	3		3
0.98	24		24	16		16
1.03	13	5	18	6		6
1.08	7	6	13	7	3	10
1.13	3	8	11	2	4	6
1.18	1	1	2		6	6
1.23		0	0			
1.28		4	4			
Total	94	24	118	48	13	61
Mean	0.94	1.12		0.95	1.14	

Two plantings, one in the field and the other in the greenhouse, show a decided association of the ovate shape and *P* gene. No peach-type fruits with ovate shape were produced in a total of 179 F_2 individuals. Using the low point in the curve between the first and second modes (see fig. 1) as a separation point, the F_2 data of Table 7, might be grouped in a dihybrid distribution as follows:

106 *PO* : 36 *Po* : 37 *pO* : 0 *po*

This indicates an appreciable linkage between the two pairs of genes, and the intensity of the linkage could be determined. From previous experience with the ovate gene, however, the writer has become skeptical of any accurate linkage values calculated from F_2 data alone, particularly in the repulsion phase. MacArthur (*5*) gives values of 17 and 8 per cent crossing over between these genes (using the pyriform type). Experiments are now in progress, involving the three genes *d–p–o* in a three-point test that will give a better measure of this linkage, and will establish accurately the order of the genes in this first chromosome.

RELATION OF OVATE SHAPE TO FRUIT SIZE

That there is a distinct association between the ovate shape of tomato fruits and their size has already been pointed out (*3*). It has also been demonstrated that the first chromosome, bearing the dwarf and peach genes, carries major size factors governing fruit size or weight (*3*). Having located the ovate gene in this same chromosome, it becomes of considerable interest to present additional data on the genetic relations between this gene and fruit size.

Fruit size in this report is given by the average weight of the same number of fruits per plant as was used in determining the shape indices from the measurements of the polar and equatorial diameters. An average of eight mature fruits of normal character were weighed individually on balance scales, readings being taken to the nearest gram.

The simple correlations between fruit shape and fruit weight of the various crosses involved in this study were as follows:

F_2—Yellow Plum × New Globe ..... $r = 0.54 + 0.04$
F_2—Yellow Plum × Dwarf Stone ..... $r = .56 \pm .04$
F_2—Yellow Plum × Red Peach ..... $r = .61 \pm .04$
F_1(YP × DS) × Yellow Plum ..... $r = .45 \pm .03$
F_1(YP × DS) × Red Pear ..... $r = .47 \pm .06$

It may be noted that in every case the correlation is high, positive, and statistically significant, indicating that ovate fruit shape is rather intimately associated with small size in these hybrid populations. In Tables 8 and 9 are included samples of these correlations, presented in order to give a visual demonstration of the relation between shape and size.

TABLE 8.—*Correlation table involving fruit shape and fruit weight in the F_2 generation of the Dwarf Stone × Yellow Plum cross*

Weight in grams \ Shape index	0.75	0.85	0.95	1.05	1.15	1.25	1.35	1.45	Total
15		2							2
20	1	9	7	4	3	1	1		26
25		4	7	14	11	9	2		47
30			3	5	9	13	4		34
35		1			14	10	4	1	30
40					11	5	2	1	19
45							1		1
50						1			1
Total	1	16	17	23	48	39	14	2	160

$r = 0.56 \pm 0.04$

TABLE 9.—*Correlation table involving fruit shape and fruit weight in the back-cross generation from F_1 (DS × YP) × YP and reciprocal*

Weight in grams \ Shape index	0.65	0.75	0.85	0.95	1.05	1.15	1.25	Total
10		1	5					6
15	1	14	32	11	23	5		86
20		6	39	19	31	27		122
25			8	4	16	16		44
30				1	5	9	2	17
35						1		1
Total	1	21	84	35	75	58	2	276

$r = 0.45 \pm 0.03$

In all of these crosses involving the ovate shape the parental variety (Yellow Plum) has in each case contributed small size (or weight) together with the ovate shape. Accordingly, any linkage with size factors located on the first chromosome would ordinarily show a tendency for an association of large size and oblate shape as well as of small size and ovate shape. This association is the one so strongly exhibited by the five correlation coefficients listed above. There is, accordingly, some evidence of linkage between shape and size factors in these crosses.

The critical test, however, will be to demonstrate that the reverse correlation (large size associated with ovate shape) exists in certain crosses of large ovate by small oblate varieties. It seems, however, that there are no commercial, large-sized tomato varieties with distinctly ovate fruits. This may be a pointed suggestion that large size is not compatible with ovate shape from the morphological standpoint, in which case the correlation between ovate shape and size noted above is not due to genetic linkage. On the other hand, a genetic linkage of this sort may be the very thing that has prevented the plant breeder from producing such a type in times past. Search for large-fruited ovate types must now be made for a critical test of the size-shape correlation. As the evidence now stands, it is merely circumstantial in pointing to a genetic linkage between ovate shape and size, due to the location of the controlling factors in the same first chromosome of the tomato.

INHERITANCE OF ROUND AND OBLATE FRUIT SHAPES

Having determined that the ovate fruit shape in tomatoes may reasonably be determined by a single, genetic factor, at least when acting in contrast both with the round (or roundish-oblate) and the oblate shapes, it becomes imperative to ascertain the interrelations of the round and oblate shapes themselves. It has been suggested that these various shapes of tomato fruits form a multiple allelomorphic system in their genetic behavior, a hypothesis that requires considerable verification.

Accordingly, data from a large series of crosses of round by oblate varieties have been assembled and will be discussed forthwith. The fruit shapes in these crosses were determined in a manner identical with those of the preceding crosses.

TABLE 10.—*Shape indices of crosses involving round and oblate fruit shapes*

Crosses	Oblate parent	Round parent	F_1	F_2	Grown
Bonny Best×Yellow Cherry	1.38	1.09	1.11	Figure 3	Greenhouse.
Golden Beauty×Red Cherry	1.50	1.06	1.15	Figure 4	Field.
New Globe×Yellow Cherry	1.29	1.07	1.09	Figure 5	Do.
Bonny Best×Yellow Peach	1.32	1.09	1.26	Figure 6	Do.
Dwarf Giant×Yellow Peach	1.35	1.09	1.27	Figure 7	Greenhouse.
Dwarf Stone×Yellow Peach			1.23	Figure 8	Do.

Six crosses involved in this investigation of the round-oblate situation have been arranged in Table 10, which gives the shape indices of the parents, F_1 and F_2 generations. In all cases samples of the three generations of plants in each cross were grown during the same season, and hence the indices are strictly comparable. The details of the F_2 and back-cross generations of each of these crosses are presented separately in Figures 3, 4, 5, 6, 7, and 8.

The six crosses in Table 10 may conveniently be grouped into two categories, first the crosses of the oblate by the round Cherry types and, second, oblate by the Peach (roundish-oblate) types. The Cherry varieties (Red and Yellow) bear fruits that are approximately round in shape, the average shape index being 1.07. The Yellow Peach variety is a trifle more oblate, its average index being 1.09.

There is a noticeable difference in the F_1 fruits of the two sets of crosses in the matter of dominance of size. Crosses of large oblate by small Cherry types give an F_1 plant with fruit showing a higher degree of dominance of small size (*2*) than is true in crosses of large oblate by the smaller Peach varieties. There is an analogous

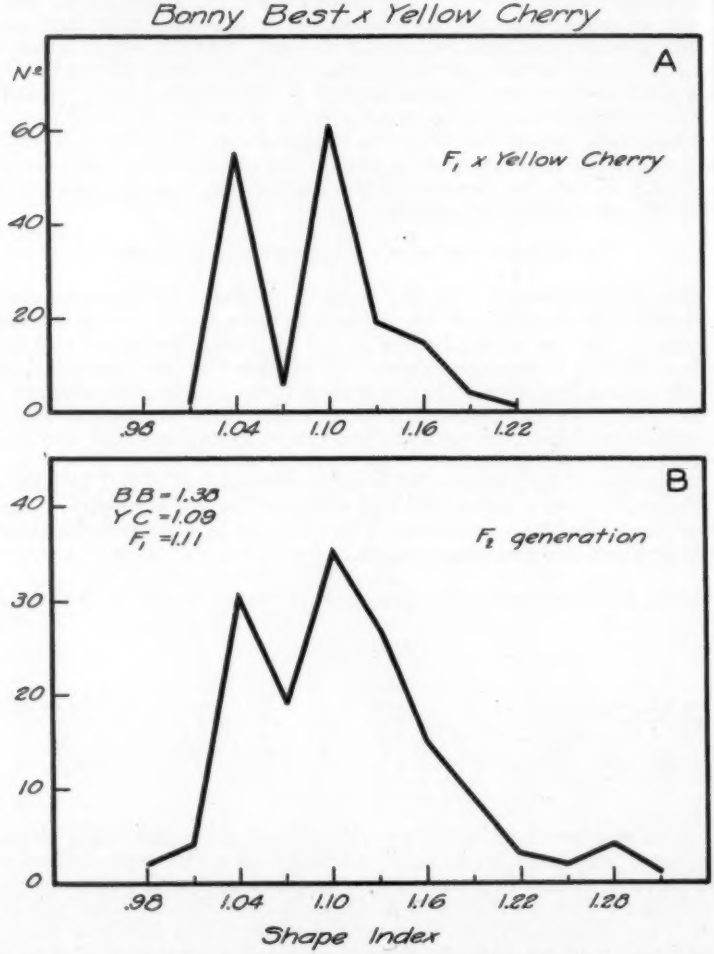

FIG. 3.—An F_2 and a back-cross generation of the Bonny Best (oblate)×Yellow Cherry (round) cross, showing distribution of fruit shape

behavior in the matter of fruit shape, the F_1 of the Peach-oblate crosses showing relatively more oblateness than the F_1 of the Cherry-oblate crosses (Table 10).

In Figure 3, showing the F_2 and back-cross generations of the Bonny Best (oblate) × Yellow Cherry (round) cross, there is definite evidence

of segregation of fruit shape. This is particularly true in the back-cross generation ($F_1 \times$ Yellow Cherry), which presents a sharp bimodal curve, indicating two major genotypes. A similar situation occurs in Figure 4, although here the differences are not so distinct.

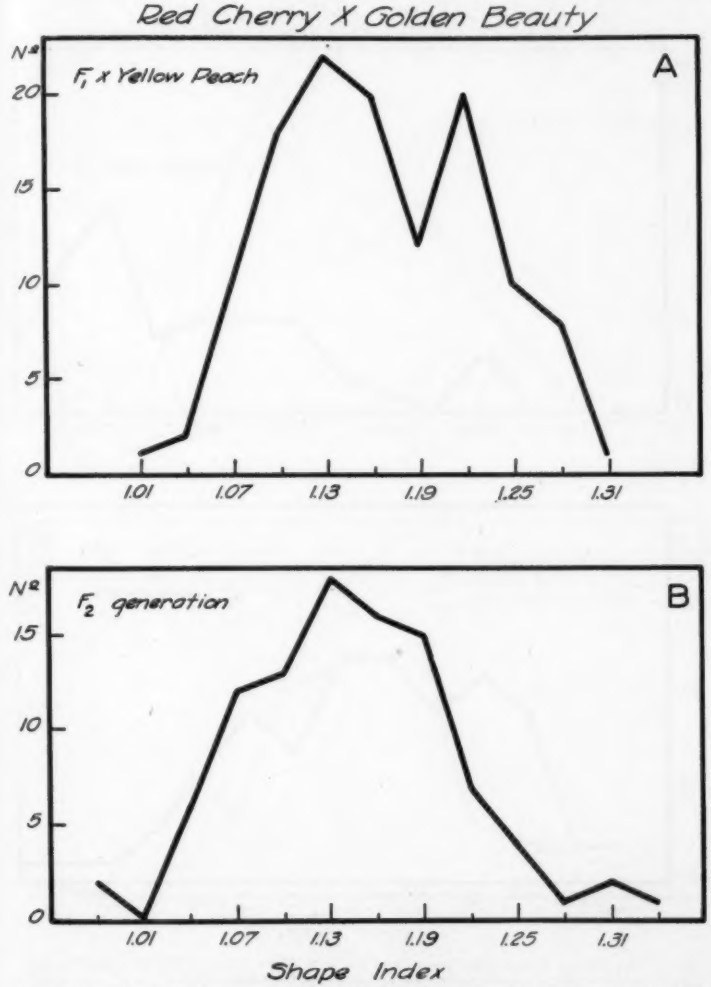

FIG. 4.—An F_2 and a back-cross generation of the Red Cherry (round) $\times$ Golden Beauty (oblate) cross, showing distribution of fruit shape

This is a cross of the Red Cherry $\times$ Golden Beauty (oblate) varieties. In this case the back cross was not made to a Cherry type but rather to the more roundish-oblate Yellow Peach variety. This might tend to decrease the influence of any two different genotypes to some extent. The third cross involving the Cherry type of fruit

in contrast with the oblate type (New Globe) appears in Figure 5. Here the F_2 generation exhibits a distinct trimodal condition. The back-cross generation to the very oblate type (Golden Beauty) shows but slight tendency toward bimodality. However, the number of

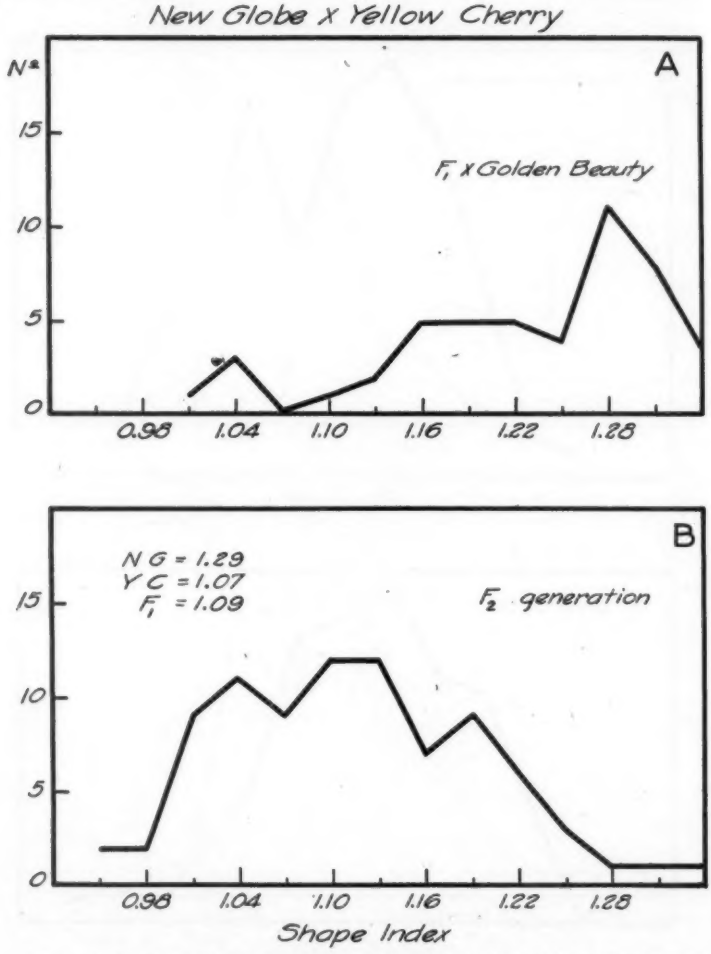

FIG. 5.—An F_2 and a back-cross generation of the New Globe (oblate)×Yellow Cherry (round) cross, showing distribution of fruit shape

plants in this back cross is really too small to offer evidence of any reliability.

Turning to the three crosses of the Yellow Peach variety (round to roundish-oblate) with the more oblate types, there is plotted in Figure 6 the first set of crosses. Here the F_2 generation (Bonny Best×Yellow Peach) exhibits some faint indications of segregation,

and the back-cross generation ($F_1 \times$ Yellow Peach) somewhat more evidence of it. Two fairly distinct modes are apparent, but they could not be classed as reliable indicators of simple genetical differences. There is better evidence in Figure 7, which shows the F_2 and back-

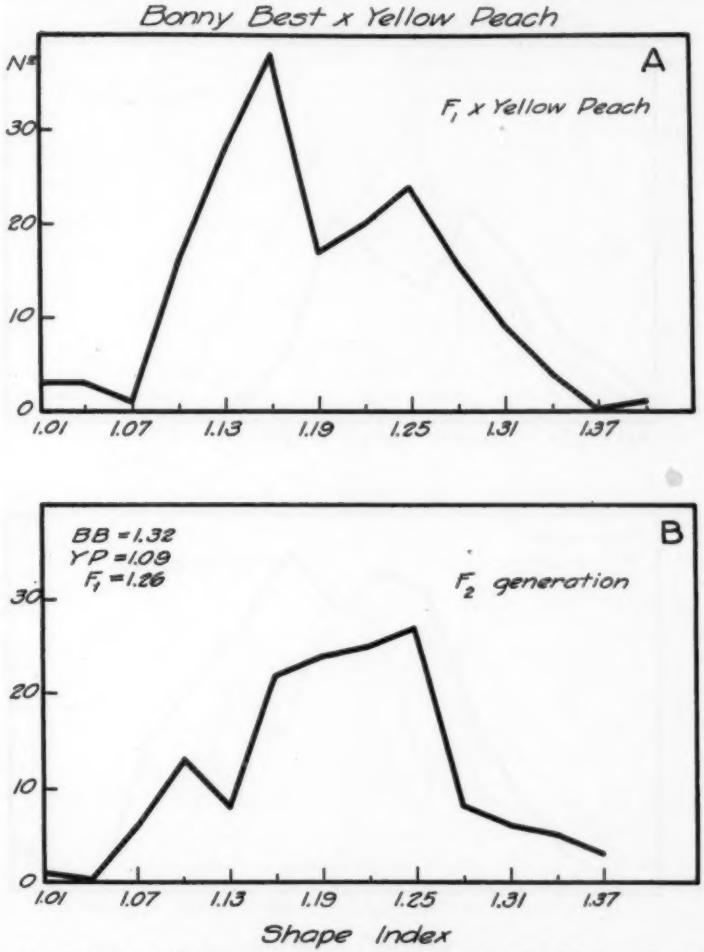

FIG. 6.—An F_2 and a back-cross generation of the Bonny Best (oblate)×Yellow Peach (round) cross, showing distribution of fruit shape

cross generations of the same Yellow Peach variety crossed with the oblate, Dwarf Giant. The back-cross generation especially is distinctly bimodal. In a similar cross, Dwarf Stone×Yellow Peach, shown in Figure 8, there is further verification of an analogous shape segregation in both F_2 and back-cross generations.

Thus in six different crosses of oblate by round types of fruit, there is some consistency in a tendency toward a simple segregation into oblate and round fruit shapes, modified, of course, by the inevitable agencies of unknown nature, mostly environmental per-

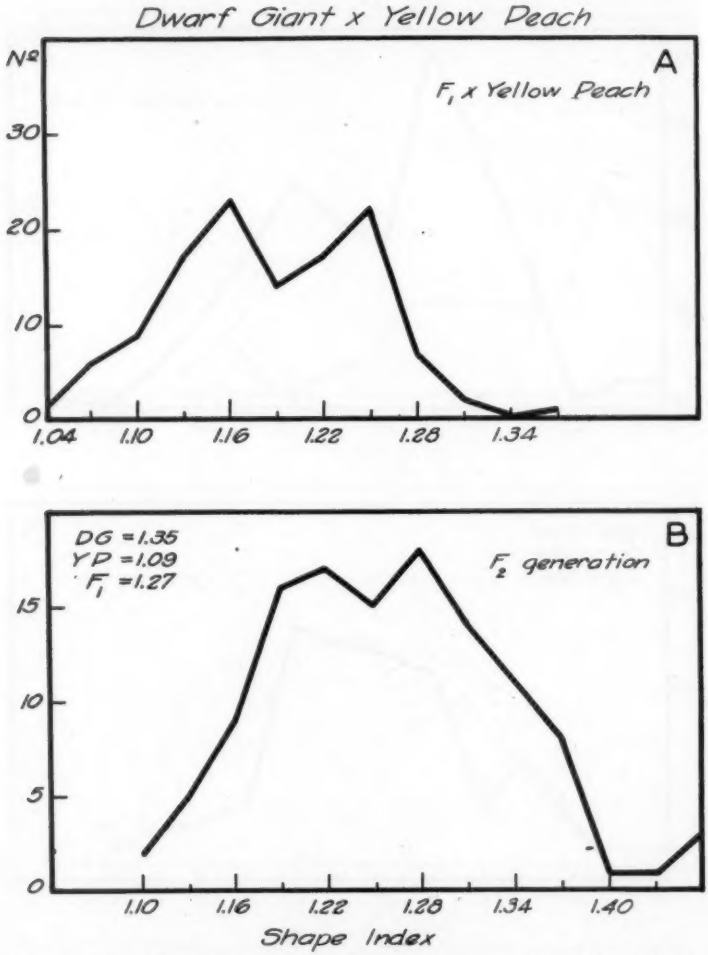

FIG. 7.—An F_2 and a back-cross generation of the Dwarf Giant (oblate)×Yellow Peach (round) cross, showing distribution of fruit shape

haps. True, in none of the crosses are the differences as marked as when the ovate shape of fruit is involved. Nevertheless, it is possible to predicate a genetic agency controlling fruit shape in the oblate-round end of the fruit-shape series that is analogous to the one discovered in the other (more recessive) end of the series. At least there seem to be major factors differentiating oblate and round shape.

A survey of the genetical relations of the oblate and round shapes of tomato fruits having suggested that it is possible for the difference between the two shapes to be attributed to a single major pair of genes, it becomes essential to consider if these genes are allelomorphic

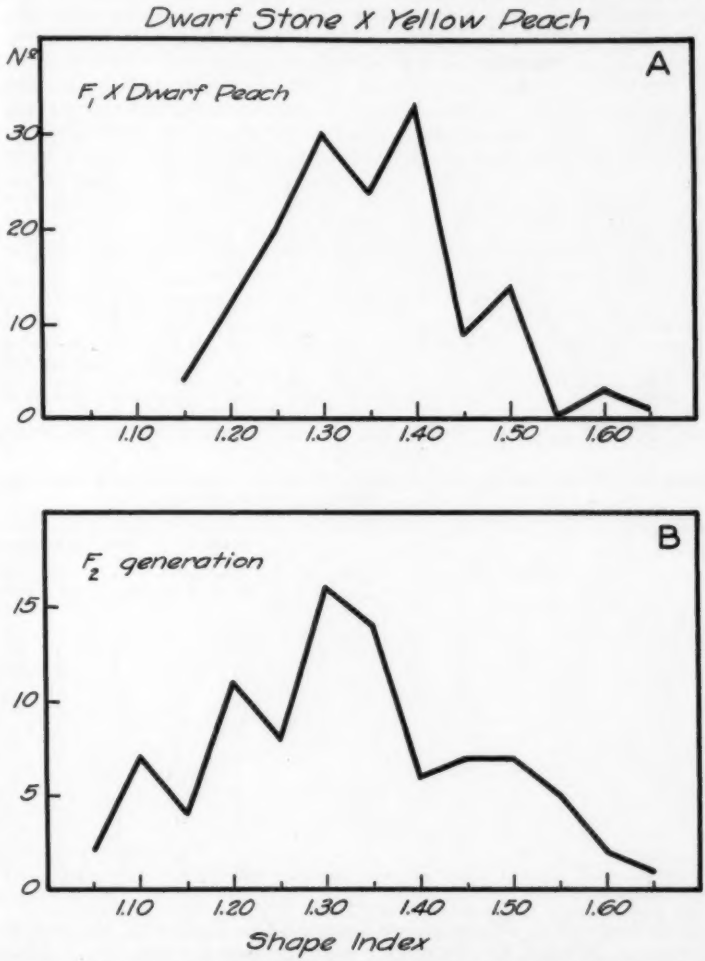

FIG. 8.—An F_2 and a back-cross generation of the Dwarf Stone (oblate)×Yellow Peach (round) cross, showing distribution of fruit shape

to the ovate genes. It will be recalled that the ovate gene has been proved to be a simple allelomorph to the oblate and also to the round-determining shape factors in crosses of oblate×ovate, and round×ovate types. This suggests strongly a multiple allelomorphic situation, but there is the other possibility that the oblate-round gene

differences discovered in the preceding crosses may be due entirely to a different set of genes than those allelomorphic to ovate shape.

TABLE 11.—*Shape indices in the F_2 and back-cross generations of the Bonny Best×Yellow Peach cross*

Shape index	F generation			F_1×Yellow Peach		
	P	*p*	Total	*P*	*p*	Total
1.01	1		1		3	3
1.04	0		0		3	3
1.07	3	3	6	1	0	1
1.10	8	5	13	3	13	16
1.13	5	3	8	14	14	28
1.16	16	6	22	18	20	38
1.19	19	5	24	11	6	17
1.22	16	9	25	12	8	20
1.25	23	4	27	14	10	24
1.28	7	1	8	11	5	16
1.31	5	1	6	3	6	9
1.34	4	1	5	2	2	4
1.37	0	0	0	0	0	0
1.40	1	0	1	0	0	0
1.43		1	1	0	0	0
1.46		0	0	1		1
1.49		1	1			
Total	108	40	148	90	90	180
Mean	1.21	1.20		1.20	1.18	
Closeness of fit	*P*=0.94		*P*=0.20			

TABLE 12.—*Shape indices of the F_2 generation of the Dwarf Giant×Yellow Peach cross*

Shape index	Stature		Fruit Pubescence		
	D	*d*	*P*	*p*	Total
1.10	2		1	1	2
1.13	5		3	2	5
1.16	9		6	3	9
1.19	13	3	8	8	16
1.22	10	7	15	2	17
1.25	9	6	11	4	15
1.28	14	4	10	8	18
1.31	10	4	8	6	14
1.34	10	1	7	4	11
1.37	4	4	6	2	8
1.40	1		1		1
1.43	1		1		1
1.46	1		1		1
1.49	1		1		1
1.52	1		1		1
Total	91	29	80	40	120
Mean	1.26	1.27	1.26	1.25	

Data bearing on this point are assembled in Tables 11, 12, and 13, which also involve the dwarf (*d*) and the peach genes (*p*). These genes belong in the first linkage group, being closely linked with the ovate genes. If the major shape differences in the oblate×round crosses are allelomorphic to the ovate gene, linkage between shape and the peach, or shape and the dwarf characters, should be evident. And the intensity of the linkage should be identical with that of the ovate gene with these characters.

In Table 11, which shows the shape distribution for the smooth (*P*) and peach (*p*) type fruit separately in the Bonny Best×Yellow

Peach cross, there is no reliable proof of any distinct linkage between fruit shape and the *Pp* genes. In the F_2 generation the smooth (*P*) fruits average 1.21 and the peach (*p*) 1.20 in shape index. The latter should give a smaller index on the linkage hypothesis, but the actual difference is doubtfully significant. If the two F_2 distributions are compared to determine whether they may be samples of the same population, the probability is extremely high ($P=0.94$) that they are identical. The same conclusion is apparent in the back-cross generation where the probability, while not so high ($P=0.20$), is sufficiently so to indicate identity. Here also there is a faint suggestion, however, that the peach fruits have a smaller index (1.18 as compared with 1.20), but this difference also is too small for a linkage that should be as close as that shown in Table 7.

These conclusions are verified in the F_2 generation of the Dwarf Giant × Yellow Peach cross in Table 12, where the average shape indices of the smooth and peach types differ only by 0.01 (1.26 − 1.25). In this cross the tall-dwarf (*Dd*) genes also are involved, and they should exhibit the same linkage (or lack of it), since they are very closely linked with the *Pp* genes. Here the dwarf fruits have an index of 1.27 and the tall types an index of 1.26. A linkage relation should give a higher index for these dwarf segregates, but the difference found is again too small to mean a great deal.

TABLE 13.—*Shape indices in the F_2 and back-cross generations of the Dwarf Stone × Yellow Peach cross*

Shape index	F_2 generation					F_1 × Dwarf Peach				
	D	*d*	*P*	*p*	Total	*D*	*d*	*P*	*p*	Total
1.00	1		1		1					
1.05	1		1		1					
1.10	7		6	1	7					
1.15	3	1	3	1	4	2	2	1	3	4
1.20	10	1	10	1	11	8	4	4	8	12
1.25	7	1	5	3	8	8	12	10	10	20
1.30	14	2	12	4	16	14	16	16	14	30
1.35	11	3	9	5	14	11	13	13	11	24
1.40	6	0	4	2	6	17	16	17	16	33
1.45	6	1	6	1	7	5	4	4	5	9
1.50	6	1	4	3	7	10	4	4	10	14
1.55	5		3	2	5	0	0	0	0	0
1.60	2		2		2	2	1	1	2	3
1.65	1		1		1	1			1	1
Total	80	10	67	23	90	78	72	70	80	150
Mean	1.32	1.32	1.31	1.35		1.36	1.34	1.34	1.35	
Closeness of fit						$P=0.80$		$P=0.81$		

In Table 13, which gives the F_2 and back-cross generations of the Dwarf Stone × Yellow Peach cross, there is a similar lack of distinct evidence of any linkage between the *Dd* or the *Pp* genes and any factors controlling oblate and round fruit shapes in this cross. The closeness of fit has been determined in the back-cross generation, and the high values of *P* (0.80 and 0.81) show that the *D* and *d*, or the *P* and *p* distributions may be considered as samples of the same population. Accordingly, there is no marked evidence of any linkage of these genes and oblate or round fruit shape as the data are presented.

Before drawing such a conclusion, however, one very important point must be considered. In all of the crosses used as evidence (Tables 11, 12, 13) it has already been shown (*3*) that these genes *Dd* and *Pp* are very closely linked with a major factor for fruit size. It has also been demonstrated that in these very three crosses involving the Yellow Peach variety, the smaller Yellow Peach variety is carrying a major factor for large size, so that the peach segregates in these hybrid generations show an average fruit weight or size considerably in excess of the smooth fruited plants. This linkage will, of course, modify the shape relations in these three crosses, particularly in view of the high positive correlation between fruit shape and fruit size. Thus the peach types, which ordinarily should bear much the smaller shape index (because the Yellow Peach parent contributed the smaller shape index), show a shape index only a trifle smaller than the smooth fruits. Likewise there is so tight a linkage between these *Pp* genes and the *Dd* factors, that this would influence the dwarf segregates in a similar manner. Accordingly, any linkage between shape and size will be obscured by this complex situation. Here is a good example of how direct experimental evidence may be misleading at times if all pertinent facts are not taken into consideration.

Unfortunately, most of the writer's crosses involving oblate and round shapes and the *Dd* or *Pp* genes were in such combinations that the unusual linkage between *D* and *P* genes and size factors would obliterate the point in question. There was one cross, however, that did not involve the peach gene. This was a cross of the Dwarf Giant (oblate) × Yellow Cherry (round). In this case the dwarf gene was contributed by the parent with the oblate fruit shape. Only 77 F_2 plants of this cross were grown. Their fruit shape indices in relation to the *Dd* genes are arranged in Table 14.

TABLE 14.—*F_2 generation from the Dwarf Giant (oblate) × Yellow Cherry (round) cross, showing linkage between fruit shape and growth habit*

Shape index	*D*	*d*	Total	Shape index	*D*	*d*	Total
1.07	1		1	1.31	1	1	2
1.10	5		5	1.34		1	1
1.13	13		13	1.37		2	2
1.16	13		13	1.40		2	2
1.19	9	1	10				
1.22	16	3	19	Total	66	11	77
1.25	5	1	6	Mean	1.18	1.30	
1.28	3	0	3				

In this cross of an oblate and a round type there is a distinct association between oblate shape and the dwarf character, as well as between round shape and the tall or standard character. This would seem to indicate that the factors controlling the difference between the oblate and round fruit shapes in this cross were linked with the *Dd* genes. Crosses like this and including other varieties with similar shape differences should give critical evidence for the hypothesis that such shape genes are allelomorphic to the ovate shape gene; in other words, that the oblate, round, and ovate shapes are determined by multiple allelomorphs.

If further proof is adduced by means of such crosses, showing that the genetic difference between oblate and round shape is caused by a pair of genes that are linked with either the *Dd* or the *Pp* genes, the chain of evidence on the multiple allelomorphic nature of the shape factors will be complete. As the matter now stands the evidence is circumstantial, with but one line of direct proof in favor of the multiple allelomorphic system. Crosses have been made that should afford critical information on this problem in the near future.

Attention should be called to the significant correlation between shape and size in the series of oblate-round crosses, a situation that is surprisingly similar to that in the ovate-oblate crosses. There follows a list of simple correlations between fruit shape and fruit size:

F_2—Bonny Best × Yellow Cherry	$r = 0.47 \pm 0.05$
F_2—Golden Beauty × Red Cherry	$r = .45 \pm .05$
F_2—New Globe × Yellow Cherry	$r = .66 \pm .04$
F_2—Bonny Best × Yellow Peach	$r = .58 \pm .04$
F_2—Dwarf Giant × Yellow Peach	$r = .52 \pm .05$
F_2—Dwarf Stone × Yellow Peach	$r = .45 \pm .05$

In these crosses of oblate by round varieties the correlations of shape and size are approximately as high as were those in the hybrid generations of the ovate by oblate varieties. This certainly points to a genetic similarity between the three shapes, the oblate, the round, and the ovate, in their reaction to fruit size, and confirms the suspicion that the three form a multiple allelomorphic series.

DISCUSSION

Fruit shape in the tomato apparently is determined by a simpler genetic mechanism than is fruit size. In these investigations certain major genes for such fruit shapes as oblate, round, ovate (including pyriform) have been isolated by quantitative means, using careful measurements of the polar and equatorial diameters of the fruit. While the effect of these genes is modified by other agents to some extent, certainly they are not as subject to environmental influence as are fruit-size factors. The segregation of the various shapes in hybrid generations proved to be unexpectedly distinct. Whereas in crossing large by small fruited types, it has been impossible to recover the large parental size in F_2 generations of 200 to 300 plants, this has been done repeatedly with respect to shape characters. Ordinarily both parental shapes are recovered in F_2, indicating a much simpler genetic basis for these shape characters.

Experimental data demonstrate rather conclusively that ovate and round shapes as well as ovate and oblate shapes are allelomorphic on a simple monohybrid basis. Both F_2 and back-cross generations give evidence of this simple relation when shape is quantitatively determined. The recessive (both round and oblate being partially dominant) ovate gene is shown by critical tests to be linked with the dwarf gene, and accordingly belongs in the first linkage group of the tomato, being removed from the dwarf gene by approximately 10 or 11 units. It is also linked with the first chromosome peach gene. This means that these allelomorphs of the ovate gene, namely, the oblate and the round determining factors, all reside in the first chromosome.

Evidence is presented to show that a very high positive correlation exists between shape and fruit size in crosses of large oblate × small ovate, or large round × small ovate types. This may be taken to mean that the ovate gene (and its allelomorphs) is closely linked with a major factor for fruit size located in the first chromosome. Previous experiments have proved that a major size factor is present in the first linkage group, there being good evidence of linked inheritance between fruit size and the *Dd* and *Pp* genes (*3*). The association of fruit size and the ovate gene seems to be corroborative testimony to the existence of definite size factors in the chromosomes of the tomato. There remains, however, the alternative explanation that the association of shape and size may be traceable to a morphological relation between the two characters in the cell growth of the tomato group, and that the size factors are in reality the result of the shape determiners.

Limited observations have shown that the pyriform shape acts genetically like the ovate shape in crosses involving the two forms. The genetic difference between the ovate and pear shape is not clearly established as yet, but it is known that the latter is incompletely recessive. There is no evidence that the gene determining pyriform shape belongs in the first linkage group.

Dominance in fruit shape, while existent, is far from complete. There seems to be only a slight dominance of the more oblate types. Intermediate shapes in crosses of these various forms are very conspicuous. The ovate or pyriform shapes disappear in the F_1 fruits when crossed with either round or oblate types. Crosses of oblate by round forms show very little dominance of the former. Thus in the series of fruit shapes noted above there seem to be differential degrees of dominance, traceable no doubt to the nature of cell growth and development in the maturing fruit.

While the hypothesis establishing a multiple allelomorphic system for the oblate-round-ovate series of fruit shapes has been developed from some rather tangible experimental data, it needs considerable verification, particularly with varieties other than those used herein. The decks are now cleared for action on this suggestive hypothesis, and the weak points in the situation need critical attention. Warren's (*6*) hypothesis of complementary action of two pairs of shape factors possibly is not in agreement with this new assumption. However, careful scrutiny of Warren's experimental data fails to demonstrate any adequate proof of his hypothesis. Where he expects an F_2 ratio of 9 "deep" to 7 "shallow" fruits, his data on six F_2 distributions are very unsatisfactory. Whereas the total of these six progenies agrees fairly well with a 9 : 7 ratio, the individual progenies show great variation, two of the six having deviations of more than three times their probable errors. What is more to the point, however, is the fact that the critical back crosses which should verify his hypothesis are all dismissed with the statement that great difficulty was encountered in classifying the fruits (by inspection), there being certain influences (fasciation) that interfered with the determination of shape. The complementary factorial hypothesis for fruit shape accordingly rests on an insecure basis.

If the suggestion of a multiple allelomorphic series involved in fruit shape should receive the necessary confirmation, it is urged that the symbolism for the genetic factors be considered as soon as possible

so as to avoid unnecessary confusion. Instead of the biliteral *pr* symbol, the basic gene symbols *Oo* (oblate-ovate) are suggested. The various allelomorphs may then be designated by priming the upper and lower case letters, or by giving them a descriptive superscript, as, for example: O = oblate; o^r = round; o = ovate (plum).

SUMMARY

By means of quantitative determinations of fruit shape (using as a shape index the ratio of the equatorial to the polar diameter) in the tomato, it is established that ovate fruit shape is determined genetically by a major, recessive gene. Back-cross data particularly demonstrate this fact. The dominant (incompletely) allelomorphs of the ovate shape were the oblate or the round shapes of the fruit.

Linkage determinations prove that this ovate-determining gene resides in the first chromosome of the tomato. Various linkage tests show that the dwarf and ovate genes are closely linked, there being approximately 10 or 11 per cent crossing over. The peach gene also is proved to be linked with the ovate-shape gene.

High, positive correlation is demonstrated between shape and size of tomato fruits in a series of F_2 and back-cross generations involving the various shapes. This may mean that fruit-shape factors of the first chromosome are linked with major fruit-size factors.

Crosses of oblate by round types give evidence of the action of single, major genes for these fruit shapes, although the differences are not so distinct as those differentiating ovate from either round or oblate shapes. Environmental agencies seem to affect oblate shape more than ovate shape. High correlations between shape and size were also found in the F_2 of these crosses.

Crosses involving oblate, round, and ovate shapes, having demonstrated that these three fruit shapes are determined by certain major genes residing in the first chromosome, the hypothesis is offered that the three form a multiple allelomorphic system. The order of dominance in this series is, oblate, round, and ovate.

There are many indications that fruit shape in general is determined by a simpler genetic mechanism than is the case with fruit size.

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THE GOSSYPOL CONTENT AND CHEMICAL COMPOSITION OF COTTONSEEDS DURING CERTAIN PERIODS OF DEVELOPMENT [1]

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INTRODUCTION

The recent findings of Schwartze and Alsberg (*7*),[3] showing that the gossypol content of cottonseeds tends to vary with the locality in which the seeds are grown, and is increased in those seeds which have a high oil content, has made it desirable to determine at what stage of maturity the maximum amount of gossypol is present. The writer desired to know if the gossypol accumulated in the seeds at the same time that there was an accumulation of other materials such as oil and protein, or if it formed after the seeds had fully matured and had separated from the plant. Ever since the isolation of gossypol as a toxic constituent of cottonseeds (*10*) the writer and coworkers at this station have been interested in the factors which determine the gossypol content of cottonseed meal and have recently published some of the results of this work (*4*, *5*). For obvious reasons, the gossypol content will depend primarily upon the gossypol contained in the original seeds. It is the writer's opinion that climatic or weather conditions are important factors to be considered and that such conditions might be more or less effective, depending upon the time of planting and perhaps of gathering the cotton. The amount of rainfall during the growing period of the cotton plant governs to some extent the amount of oil in the seeds, and if it can be demonstrated that the gossypol content is regulated by the formation of oil, then these same conditions will in turn affect the gossypol content. Present knowledge of the gossypol content of cottonseeds as dependent upon variety, oil content, and geographical region of growth of the seeds is confined to the publication cited above (*7*).

Balls (*2*) working with Egyptian cotton No. 77 finds that the cottonseed has reached its full size between 18 and 21 days after the flowers have opened, the boll reaching its full size soon after, but that the seed continues to develop for some time. After 45 days the boll begins to crack and the cotton is usually ready to be picked within 5 or 6 days thereafter. Garner, Allard, and Foubert (*6*) studying the oil content of cottonseeds at different stages of maturity, found that the increase of oil proceeded somewhat more rapidly than the growth of the seed. In experiments with soy beans these investigators found that during the final stages of ripening there

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[2] The writer is indebted to L. L. Ligon, in charge of cotton investigations, for much information and help in obtaining the cottonseeds, and to V. G. Heller, head of the department of agricultural chemical research, and N. B. Guerrant, assistant chemist, for their helpful suggestions and advice.

[3] Reference is made by number (italic) to "Literature cited," p. 991.

was a slight decrease both in the size of the seed and in the oil content. This phenomenon they attributed to continued respiratory activity after assimilation had ceased. Referring to the transformation of carbohydrate to fat they state that "under proper conditions this transformation takes place in unripe seeds detached from the mother plant, further indicating that the oil is derived from the carbohydrates."

Since gossypol contains only carbon, hydrogen, and oxygen, and is closely associated with the oil in cottonseeds in other ways, it appeared possible that it might also be formed at the same time and that it might be associated in some way with the formation of the oil. The practical value of any such finding is perhaps obscure until our very limited knowledge of the formation of gossypol and its properties, except as these properties are manifested in the feeding of cottonseed products to livestock, is considered. Concerning the rôle of gossypol in the life of the cotton plant little is known. Carruth (*3*) speaks of gossypol as being formed by the disintegration of adjacent cells. In the light of the histological studies upon the glands of the cotton plant by Stanford and Viehoever (*9*), this statement is well founded.

EXPERIMENTAL MATERIAL AND METHODS

COLLECTING SAMPLES

Cottonseeds of a selected variety, Oklahoma Triumph 44, were used in this study. They were obtained from the field plots in which cotton variety tests were being made by the department of agronomy. The cotton crop, grown on upland soil, was planted May 10, and picking began October 15. There was a moderate amount of rainfall throughout the growing season, and the season was favorable for a large cotton crop. On October 1 bolls at different stages of development as determined by their appearance and condition, were collected in the field and brought to the laboratory for an analysis of the seeds. In this collection were full-size bolls approximately 50 days old, and showing signs of cracking, bolls which had just opened; bolls which had been open for 2 or 3 days; those which had been open for 5 or 6 days; and finally, bolls which had been open for longer periods and from which the cotton was hanging down, the locks having lost much of their firmness.

On February 1 another collection of matured seeds was made from the unpicked cotton left in the field, and at the same time a representative sample of seeds from the whole field was obtained from the cotton gin. All the seeds were removed from the bolls by hand and delinted with sulphuric acid. The immature seeds were dried at a temperature sufficiently low to prevent the destruction of gossypol or its change to a less soluble form. The seeds were then ground separately and sampled for chemical analysis, including gossypol determinations. Representative samples of the unground seeds were reserved for determinations of their total dry matter.

The methods of analysis used by the Association of Official Agricultural Chemists (*1*) were employed for the chemical determinations, anhydrous diethyl ether being used as an extractive for the fat determinations. The gossypol was determined by Carruth's method as modified by Schwartze and Alsberg (*7*). Fifty-gram samples were used and the seeds were extracted for 24 hours in a Soxhlet extraction

apparatus. Calculation of the gossypol in the aniline gossypol precipitates was made by use of the conversion factor .74, since the composition of this precipitate approaches $C_{30}H_{28}O_9 2C_6H_5NH_2$, and this factor has been used by previous investigators (*8*).

DISCUSSION AND RESULTS

PROXIMATE ANALYSIS

The results of the several analyses are given in Table 1.

TABLE 1.—*Chemical composition and gossypol content of cottonseeds at different stages of maturity*

Sample No.	Condition of boll or seeds	Composition of seeds on dry-matter basis					
		Ash	Crude protein	Crude fiber	Nitrogen-free extract	Ether extract	Gossypol
		Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
1	About to open	6.20	28.46	18.56	32.81	13.97	0.048
2	Just open	3.93	31.11	14.46	26.49	24.01	.428
3	Open 2 to 3 days	3.16	28.46	15.13	28.43	24.82	.461
4	Open 5 to 6 days	3.15	29.86	15.33	27.93	23.73	.538
5	Open over 6 days	3.33	28.37	15.20	28.69	24.41	.546
7	Mature seeds picked Feb. 1	3.24	30.29	14.37	26.34	25.76	.551
6	Seeds obtained from gin	3.34	29.01	15.78	28.46	23.41	.452

From these results it would seem that there is a period of intense oil formation occurring about the time the boll begins to crack and just previous to its opening. This wide difference in the oil content of the seeds picked at these two intervals appeared questionable, and another picking of the unopened bolls was made. The oil content of the seeds obtained in this second picking, 15.05 per cent, was only slightly higher than the one shown in Table 1, which satisfied the writer that the first figures obtained were approximately correct. Other factors not considered here may influence the oil content of the seeds at this stage; but as this study was concerned primarily with gossypol determinations, the influence of these factors was not investigated further. However, it is interesting to note that there is a slight but variable increase in the oil content of the more mature seeds and that this amount did not show a decrease even when the seeds were allowed to remain in the field for several months after they had matured. The relative amount of oil in the seeds was nearly doubled between the first and last stages of development.

The protein content of the immature and mature seeds does not show any marked increase as the seeds develop. This indicates what might be expected, namely, an early deposition of nitrogenous material. There are, however, fluctuations between the protein content of seeds picked at different times which may be accounted for by the variable size of the seeds and by errors introduced in obtaining samples. The same conditions hold true for the results of the fat determinations, although an attempt was made to eliminate these errors as far as possible by obtaining samples from a selected area of the field. At a later date the writer hopes to be able to make a more thorough study of the accumulation of both oil and protein during the successive stages of development of the cottonseed.

The ash, crude fiber, and nitrogen-free extract of seeds from opened bolls were considerably less than from unopened bolls, but remained quite constant for the seeds picked during the later stages of development. These general results are in harmony with the findings of previous investigators working with other kinds of seeds as well as with the results of some unpublished work in this laboratory by N. B. Guerrant.

GOSSYPOL DETERMINATIONS

The gossypol determinations on seeds at various stages of maturity given in Table 1 showed some very interesting results. The gossypol content of the mature seeds was much higher than the writer has found for other samples grown in this locality, which were of no particular variety and had been in storage for some time. The formation of gossypol proceeded much more rapidly than that of oil, the gossypol making its greatest increase during the first two stages and continuing to increase at a very slow rate during the later stages. Although there are some indications that gossypol formation is concurrent with the formation of oil, there seems to be no direct relationship between the two; and in the case of sample No. 4, which showed a relatively lower oil content, the gossypol content was increased. The high gossypol content of those seeds which had matured and were left standing in the field is perhaps more indicative of the lysigenous origin of gossypol.

WEIGHT OF COTTONSEED IN RELATION TO ITS CHEMICAL COMPOSITION AND GOSSYPOL CONTENT

In order to observe the absolute as well as the relative changes in the oil content of cottonseeds collected at two stages of maturity, Garner, Allard, and Foubert (*6*) determined the weight of the seed and from the weight calculated the grams of oil in 1,000 seeds. Although the changes occurring in the weight of the seeds do not necessarily represent the exact growth made by the seeds between the stated intervals, it was felt that to proceed on such an assumption might aid in interpreting the data obtained in this study.

The dry weight of 500 seeds from several pickings was determined and the analysis of these calculated from the figures in Table 1. From these results, which are given in Table 2, it is evident that there was considerable growth between the first two stages, resulting in increased amounts of all the constituents.

TABLE 2.—*Chemical composition and gossypol content of 500 cottonseeds at different stages of development*

Sample No.	Condition of boll or seeds	Weight of dry seeds	Ash	Crude protein	Crude fiber	Nitrogen-free extract	Ether extract	Gossypol
		Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
1	About to open	20.09	1.25	5.72	3.73	6.59	2.81	0.010
2	Just open	38.46	1.51	11.96	5.56	10.20	9.23	.165
3	Open 2 to 3 days	43.27	1.37	12.32	6.55	12.30	10.74	.200
4	Open 5 to 6 days	44.77	1.41	13.36	6.86	12.51	10.62	.241
5	Open over 6 days	40.63	1.35	11.53	6.18	11.65	9.92	.222
7	Mature seeds picked Feb. 1	40.32	1.31	12.25	5.80	10.58	10.38	.222
6	Seeds obtained from gin	40.96	1.36	11.88	6.46	11.66	9.60	.185

A comparison of Tables 1 and 2 serves to emphasize some important differences in the manner of presenting data of this kind.

From Table 1 it appears that there is a decrease in the crude fiber, ash, and nitrogen-free extract of cottonseeds as they mature, while Table 2 shows an increase in the constituents. Both tables show an increase in the amounts of the other constituents which represent the greatest changes taking place in the seeds as they develop up to the time that the boll has been open for several days. Following this period the results are more or less irregular. The writer believes Table 2 to be a more correct interpretation of what actually happens in the seeds and has used the figures given in this table as a basis for drawing his conclusions. No explanation is offered for the large difference in weight between the seeds in samples Nos. 4 and 5 at this time, although a slight decrease in weight may be expected, due possibly to respiration. The gossypol content of the seeds is shown to be quite independent of their weight, but is slightly increased with the age of the seeds; and these facts lead the writer to think that storage conditions may also have an effect upon this amount.

SUMMARY

The gossypol content and chemical analyses of cottonseeds of the variety Oklahoma Triumph 44, picked at different stages of development, are reported. The seeds were obtained at the different stages from the time the boll was ready to open until it had fully opened, and also included those seeds which had remained in the field unpicked for several months. The greatest change in the composition of the seeds over the range studied occurred at the time the boll was mature and about to crack, from which time until the boll opened the gossypol content increased rapidly and continued to increase for some time thereafter. This increase in gossypol was greater than the increase of any of the other constituents, all of which showed only small increases after the boll had opened. No correlation was found between the formation of oil and the formation of gossypol.

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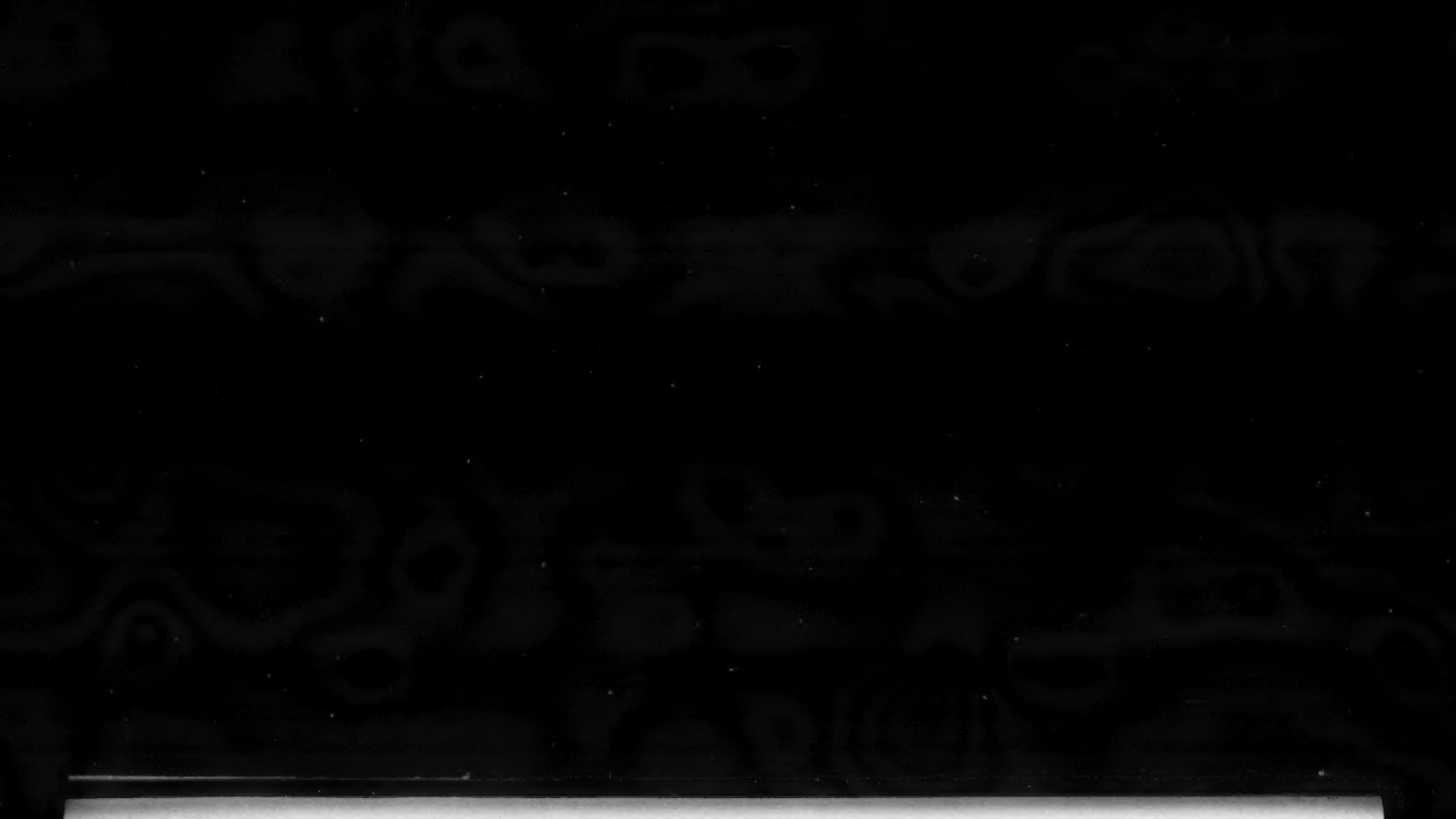
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